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MORPHOLOGICAL AND PHYSIOLOGICAL VARIABILITY OF SPECIES OF *MELOIDOGYNE* IN WEST AFRICA AND IMPLICATIONS FOR THEIR CONTROL

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MORPHOLOGICAL AND PHYSIOLOGICAL VARIABILITY OF SPECIES OF *MELOIDOGYNE* IN WEST AFRICA AND IMPLICATIONS OF THEIR CONTROL

(with a summary in Dutch)

PROEFSCHRIFT

TER VERKRIJGING VAN DE GRAAD
VAN DOCTOR IN DE LANDBOUWWETENSCHAPPEN
OP GEZAG VAN DE RECTOR MAGNIFICUS
DR. H. C. VAN DER PLAS
HOOGLERAAR IN DE ORGANISCHE SCHEIKUNDE
IN HET OPENBAAR TE VERDEDIGEN
OP DONDERDAG 12 JANUARI 1978
DES NAMIDDAGS TE VIER UUR IN DE AULA
VAN DE LANDBOUWHOGESCHOOL TE WAGENINGEN

H. VEENMAN & ZONEN B.V. – WAGENINGEN – 1978

STELLINGEN

1

Het vaststellen of een bepaalde plant al dan niet een geschikte waardplant is van een in de grond voorkomende *Meloidogyne* populatie kan alleen geschieden door experimenten.

2

De teelt van plantesoorten welke geen waardplant zijn van wortelknobbelaaltjes en van rassen van gewoonlijk vatbare gewassen moet tegen deze nematoden voornamelijk preventief worden toegepast.

3

In tegenstelling tot de gangbare opvattingen kunnen larven van wortelknobbelaaltjes zich in de grond in korte tijd over meer dan 40 cm verplaatsen.

4

Bij de beschrijving van nieuwe nematodensoorten dienen zo mogelijk ook cytologische en fysiologische gegevens te worden vermeld.

5

Quarantainevoorschriften van verscheidene landen zijn niet realistisch omdat zij onvoldoende rekening houden met de fysiologische specialisatie van de schadeverwekkers.

6

Mocht het mogelijk zijn het nif-complex te incorporeren en te laten functioneren in gramineeën, dan zou dit een belangrijke bijdrage van de moleculaire biologie aan de landbouw zijn.

7

Landbouwvoorlichting in achtergebleven gebieden heeft weinig zin voordat de aanvoer en beschikbaarstelling van voor de landbouw benodigde materialen en de afvoer van het geoogste product zijn verzorgd.

8

Een bestrijdingscampagne tegen ingewandsparasieten in een achtergebleven gebied kan als indirect gevolg hebben dat het aantal gevallen van open longtuberculose in zo'n gebied sterk daalt.

9

Er bestaat een grote overeenkomst tussen de vocale en instrumentale vertolkingen van psalmen door de Harristen en de eerste geregistreerde muziek van de Amerikaanse negers.

C. NETSCHER
Wageningen, 12 januari 1978

VERANTWOORDING

Vanaf oktober 1960 tot juli 1962 was de auteur werkzaam als phytopatholoog aan de University of Science and Technology te Kumasi, Ghana. Gedurende deze periode kwam hij voor het eerst in aanraking met nematologische problemen in de landbouw, vooral die veroorzaakt door wortelknobbelaaltjes (*Meloidogyne* spp.).

Sinds augustus 1962 is hij werkzaam als nematoloog bij het Office de la Recherche Scientifique et Technique Outre-Mer (ORSTOM), de Franse regerings-organisatie voor onderzoek in de Tropen, eerst in Adiopodoumé (Ivoorkust) en vanaf 1970 in Dakar (Senegal). Deze periode werd onderbroken door een aantal korte verblijven in Nigeria, Mauretanië, Upper-Volta, Indonesië, Engeland, Nederland, Canada en de Verenigde Staten, gedurende welke gespecialiseerde kennis omtrent het geslacht *Meloidogyne* werd opgedaan resulterend in een aantal verslagen en artikelen omtrent de plantenparasieten die tot dit geslacht behoren.

Dit proefschrift is een synthese van deze artikelen, andere die betrekking hebben op fundamenteel nematologisch onderzoek, technieken en veldproeven, alsmede van nog niet eerder gepubliceerde gegevens en waarnemingen.

Hieronder volgt een lijst van publicaties welke mede de basis van dit proefschrift vormen:

1. NETSCHER, C. (1970). a. Les nématodes parasites des cultures maraîchères au Sénégal. Cah. ORSTOM, Sér. Biol. **11**: 209–229.
2. NETSCHER, C. (1970). b. A rapid technique for mass-killing of nematodes with hot fixative. Nematologica **16**: 603.
3. NETSCHER, C. (1973). Étude sur la variabilité de la longueur des larves chez *Meloidogyne incognita* Chitwood, 1949, et *Meloidogyne javanica* Chitwood, 1949. Cah. ORSTOM, Sér. Biol. **21**: 91–95.
4. NETSCHER, C. (1974). L'arachide et le contrôle biologique des nématodes *Meloidogyne* spp. dans les cultures maraîchères du Sénégal. C. r. hebd. Séanc. Acad. Agric. Fr. **60**: 1332–1339.
5. NETSCHER, C. (1975). Studies on the resistance of groundnut to *Meloidogyne* sp. in Senegal. Cah. ORSTOM, Sér. Biol. **X**: 227–232.
6. NETSCHER, C. (1977). Observations and preliminary studies on the occurrence of resistance-breaking biotypes of *Meloidogyne* spp. on tomato. Cah. ORSTOM, Sér. Biol. **XI**, 1976: 173–178.
7. NETSCHER, C. & LUC, M. (1974). Nématodes associés aux cultures maraîchères en Mauritanie. Agron. trop., Nogent **29**: 697–701.
8. NETSCHER, C. & MAUBOUSSIN, J. C. (1973). Résultats d'un essai concernant l'efficacité comparée d'une variété de tomate résistante et de certains nématicides contre *Meloidogyne javanica*. Cah. ORSTOM, Sér. Biol. **21**: 97–102.

9. NETSCHER, C. & PERNES, J. (1971). Étude concernant l'influence de la constitution génétique sur la longueur des larves d' *Heterodera oryzae*. Nematologica **17**: 336–346.
10. NETSCHER, C. & SEINHORST, J. W. (1969). Propionic acid better than acetic acid for killing nematodes. Nematologica **15**: 286.
11. TAYLOR, D. P. & NETSCHER, C. (1974). An improved technique for preparing perineal patterns of *Meloidogyne* spp. Nematologica **20**: 268–269.
12. TAYLOR, D. P. & NETSCHER, C. (1975). Occurrence in Senegal of a biotype of *Meloidogyne javanica* parasitic on strawberry. Cah. ORSTOM, Sér. Biol. **X**: 247–249.

CONTENTS

1. INTRODUCTION

Members of the genus *Meloidogyne* (Goeldi, 1887) Chitwood, 1949, are called 'root-knot' nematodes because they induce galls on the roots of most plants attacked. Species of *Meloidogyne* cause the most important nematological problem in agriculture of developing countries in the tropics for the following reasons: many tropical crops are heavily damaged by species of *Meloidogyne*; disease complexes exist in which root-knot nematodes increase the severity of important fungal and bacterial diseases (e.g. *Fusarium* wilt of tomato and bacterial wilt of tobacco), and these nematodes are widespread and frequently abundant in tropical soils.

Though few reliable data exist on losses caused by *Meloidogyne*, the following examples illustrate the economic importance of these nematodes. In South Carolina, USA, severe attacks of *M. incognita* (Kofoid & White, 1919) Chitwood, 1949, on potato caused losses up to $ 1000 per acre (SITTERLY & FASSULIOTIS, 1965). Cotton yields increased 50% following nematicide treatment of infested land (RASKI *et al.*, 1953). Field trials in North Carolina showed yield reductions of 85% of tomato in soils infested with *M. incognita* compared to yields of uninfested plots (BARKER *et al.*, 1976). Total crops failure was observed by MILNE (1972) in tobacco fields infested with *Meloidogyne* sp.

Contrary to most plant diseases and pests in which minute inocula can initiate epidemics under conditions favourable for the parasites, damage caused by nematodes depends on the initial population density present at the time of planting. Therefore, nematode control consists of reducing soil populations to such levels that susceptible crops can be grown without suffering great losses from the remaining nematodes.

A common method of nematode control is the application of special chemicals, nematicides, that kill a large proportion of nematodes present in the soil. Unfortunately, nematicides are expensive, toxic and usually difficult to use for persons lacking experience in pesticide application. For instance, application of most of these chemicals necessitates the purchase of rather costly equipment easily damaged by these corrosive products. In order to obtain the maximum benefits from the nematicide, manufacturers' recommendations should be strictly followed. This will avoid low nematode kill and even crop injury in the case the chemicals are phytotoxic. Therefore control methods not involving the use of nematicides are of great interest to the average farmer, especially in the tropics.

As is true for other soil-borne pathogens, crop rotations offer a possible way to control *Meloidogyne*. Like all plant – parasitic nematodes, species of *Meloidogyne* are obligate parasites that must feed on a suitable host to complete their lifecycles and to reproduce. Thus, crop sequences which include non-

hosts and resistant varieties of otherwise suitable host plants should decrease soil populations of root-knot nematodes and improve conditions for production of susceptible crops. Unlike air-borne pathogens dispersion of nematodes is limited and little or no reinfestation from nearby inoculum reservoirs will occur.

Unfortunately many complications interfere with the application of crop rotation as a control method for *Meloidogyne*. The extreme morphological and physiological variability within some species of this genus in West Africa makes reliable identification and establishment of host ranges of these species very difficult. The implications of this variability are discussed in this paper with special reference to the situation existing in West Africa.

2. TAXONOMY AND MORPHOLOGICAL VARIABILITY OF *MELOIDOGYNE*

2.1. HISTORICAL ASPECTS

Undoubtedly lack of sufficient communication between scientists during the end of the nineteenth and the beginning of this century resulted in root-knot nematodes being described under different names. WHITEHEAD (1968) reviewed the history of the genus, the existing synonyms and the lumping in 1932 of all described root-knot species under one species: *Heterodera marioni* (Cornu, 1887) Goodey, 1932.

Within the genus *Heterodera* Schmidt, 1871, the root-knot nematode occupied a very special place. It was the only species in the genus capable of inducing galls on the roots of parasitized plants, and, unlike the other species, *H. marioni* females were soft-walled and after their death did not transform into egg-containing cysts. Eggs of *H. marioni* are deposited in a gelatinous matrix attached to the posterior part of the females. Most species of *Heterodera* do not produce egg-masses although exceptions are known; for example, *H. schachtii* Schmidt, 1871, *H. oryzae* Luc and Berdon, 1961, and others.

Another important difference between the root-knot nematode and other species of *Heterodera* was the polyphagous nature of *H. marioni* compared to the relative host specificity of the other species. Several hundreds of plants belonging to many families were recorded as hosts for *H. marioni* whereas the most polyphagous of the other species, *H. schachtii*, mainly parasitizes plants belonging to the families Polygonaceae, Chenopodiaceae and Cruciferae.

Though the differences mentioned above together with certain morphological characters typical for *H. marioni* justify the separation of the root-knot nematode from the genus *Heterodera*, the species was maintained in the genus because of the general morphological resemblance to the other species. This lasted until 1949 when CHITWOOD, studying different populations of root-knot nematodes, was able to indicate morphological differences between five species and a subspecies. He reestablished the genus *Meloidogyne* Goeldi, 1887, into which he placed the newly described species; *Meloidogyne* together with the genus *Heterodera* constituted the subfamily *Heteroderinae*. WOUTS and SHER (1971) finally placed the root-knot nematodes in the subfamily Meloidogininae Skarbilovitch, 1959.

The main character CHITWOOD used to differentiate between the species of *Meloidogyne* was the so-called perineal pattern, the cuticular ornamentation around the anus and the vulva of the female formed by striae and sometimes the folds of the cuticle. He listed a few other characters, such as the shape of the stylet knobs and the length of stylets of males and females which could serve as supplemental characters to assist in the identification of certain species: how-

ever, most measurements utilized by CHITWOOD cannot be used to identify the species, as no clear-cut differences exist between species for several of these characters.

Therefore, emphasis was placed on the configuration of the perineal pattern for identifying species of *Meloidogyne*. TAYLOR, DROPKIN and MARTIN (1955) gave a key to the species of *Meloidogyne* known at that date based entirely on the characters of the perineal pattern.

Unfortunately, it was soon observed that the perineal pattern varied greatly between and within populations making identification of root-knot nematodes difficult. Almost every student of *Meloidogyne* has experienced great difficulties in making specific determinations. Aberrant forms and intermediates between species are frequently observed making identification uncertain and it has appeared that the criteria used for identification are not well-defined. Thus identification may be to some degree a subjective judgement rather than an objective determination.

The variability within root-knot species was well recognized by Chitwood himself, who stated in his original paper: 'Members of the genus *Meloidogyne* are extremely adaptable and their morphological characters show considerable variation. In fact we have not as yet seen two identical specimens. Nevertheless general pattern series and other structures show similarities, and progenies from individual females are relatively consistent both as to morphology and host range'. In the same paper he wrote: 'It would be remiss not to mention that twice we have encountered individual females in which the perineal pattern on one side of the body was that of *M. incognita* and on the other side it was in one instance *M. i. acrita* and in the other case it was *M. javanica*. Both cases occurred in mixed natural populations. Male stylets of *M. javanica* are very similar to those of *M. incognita* and of course the whole anatomy of *M. incognita* and *M. i. acrita* is very similar'. Thus, even the author of these species admitted that species identification based upon perineal patterns was not completely reliable.

ALLEN (1952) stated: 'Satisfactory identifications are made only with difficulty due to the extreme individual variation that may be present in certain populations, even those originating from the progeny of a single female'.

In the progeny of a single female collected from cotton, multiplied on tomato, and subsequently cultured on different hosts, including cotton, alfalfa and sugar beet, ALLEN found a majority of females with perineal patterns of *M. incognita acrita* Chitwood, 1949. However, certain patterns closely resembled those of *M. javanica* (Treub, 1885) Chitwood, 1949. Because of this variability, he concluded that specific identification of field populations should be based on a fairly large series of perineal patterns.

In a detailed study in which one pure line of *M. arenaria* (Neal, 1889) Chitwood, 1949, and two of *M. incognita acrita* Chitwood, 1949, each established from a single juvenile, were compared with populations of mixed ancestry of these two species, DROPKIN (1953) attempted to quantify the mor-

phological characters of the perineal patterns. Using photographs of the patterns, made with a 40 × oil immersion objective, printed at the same optical magnification, standard tracings of the patterns were made; areas obtained in this way and morphological features such as vulval width and distance from anus to vulva were determined. Analysis of the data showed that patterns of pure lines of *M. incognita acrita* were less variable than those of populations of mixed ancestry; in *M. arenaria* no differences could be demonstrated. A particular pattern ('E type') found among wild type *M. incognita acrita* occurred with high frequency in two generations of offspring of a female possessing the pattern. Although this study suggested that shape and possibly some of the details of the patterns were under genetic control, the figures presented by DROPKIN (1953) cannot serve to distinguish between *M. arenaria* and *M. incognita acrita*. In fact, for all characters measured, overlapping existed between the populations of the two species studied; for example, the mean vulval width of the populations of *M. incognita acrita* ranged from 2.3 to 4.8 ocular micrometer units, whereas that of *M. arenaria* was between 2.4 and 3.

In a study of all species of *Meloidogyne* known at that date, TAYLOR, DROPKIN and MARTIN (1955) suggested a system of nomenclature for the perineal pattern and presented a key based on its characters. They recognized that patterns vary to a greater or lesser extent within the species so that the extremes in one species could be mistaken for those of another.

TRIANTAPHYLLOU and SASSER (1960) studied females from eggmass cultures derived from different populations of *M. incognita* and *M. incognita acrita*. They synonymized the two because the perineal patterns of different lines ranged from *M. incognita* through intermediate types to *M. incognita acrita*. They confirmed the observations of DROPKIN (1953) that certain characteristics of cultures persisted from the first to the 10th or 12th generation, a phenomenon suggesting that some of the pattern characteristics were under genetic control. Though most nematologists seemed to have accepted this view (WHITEHEAD, 1968), not all agreed. Recently ESSER, PERRY and TAYLOR (1976) rejected this synonymy on the basis of statistical evidence presented by TERENTEVA (1967) and the absence of an inflated rectum in juveniles of *M. incognita acrita* as opposed to the inflated rectum of juveniles of *M. incognita*. We later will discuss the validity of this separation.

During the first ten years after Chitwood reestablished the genus *Meloidogyne*, only four new species or subspecies were described; *M. arenaria thamesi* Chitwood, 1952 in CHITWOOD, SPECHT and HAVIS, 1952; *M. brevicauda* Loos, 1953; *M. acronea* Coetzee, 1956; and *M. inornata* Lordello, 1956. However, from the beginning of the 1960's a great number of new species has been described. Thus, WHITEHEAD (1968) listed 23 species of *Meloidogyne* and one species inquirenda. Because of the unreliable nature of perineal patterns to make valid specific identifications, Whitehead used measurements and morphology of juveniles and males to elaborate a system for defining different species of *Meloidogyne* and a procedure was suggested in which up to 50 females had

to be examined. He concluded that the most useful measurements for taxonomic studies were body length and tail length of the juveniles (i.e. the second-stage larvae), stylet length in males and females and certain measurements of the perineal patterns such as vulval width, interphasmidial distance, distance from anus to vulva, and distance from anus to tail terminus. Morphological characters most appropriate for identifying root-knot species were in the males: position of the cephalids, appearance and annulation of the head, form of stylet knobs, position of phasmids and form of the spicules; in the females: the perineal pattern, and in the juveniles: the appearance of the head and the form of the tail.

SLEDGE and GOLDEN (1964) created a new genus, *Hypsoperine*, close to *Meloidogyne* but differing from the latter by its thickened cuticle and an elevated perineal area of the females. This genus was synonymized with *Meloidogyne* by WHITEHEAD (1968) because he found these features independantly in several species of *Meloidogyne*.

Since WHITEHEAD's important paper additional species have been described and, recently, ESSER, PERRY and TAYLOR (1976) published a diagnostic compendium for the identification of 35 root-knot nematode species. They formulated three lattices, one each for females, males, and juveniles. In each lattice characters were presented, considered essential for the identification of species of *Meloidogyne*.

2.2. CYTOLOGICAL ASPECTS

TRIANTAPHYLLOU (1962, 1963, 1966, 1969, 1971a, b) studied the cytology of several species of *Meloidogyne*. He established that *M. arenaria*, *M. incognita* and *M. javanica* reproduce by mitotic parthogenesis. Each of these species is characterized by a different degree of polyploidy; within each species chromosome numbers of different populations vary within a certain range, suggesting different degrees of aneuploidy of these populations. *M. hapla* Chitwood, 1949: *M. graminicola* Golden and Birchfield, 1965; *M. naasi* Franklin, 1965: *M. graminis* (Sledge and Golden, 1964) and *M. ottersoni* (Thorne, 1969) are all characterized by a meiotic parthenogenetic mode of reproduction. When males were abundant amphimictic reproduction was observed in *M. hapla*, *M. graminis*, *M. graminicola*, and *M. ottersoni*. It was suspected that amphimixis might occur in *M. naasi*. Certain populations of *M. hapla* have a mitotic type of parthenogenetic reproduction. Of all species of *Meloidogyne* studied to date, only *M. carolinensis* Fox, 1967*, has an obligate amphimictic reproduction. Though of great value in determining phylogenetic relationships between different species of *Meloidogyne*, up till now cytological techniques have had little value for specific determination. Accurate counts are difficult to make because

* *M. carolinensis* is not an officially recognized species and it has not technically been published.

of the great number of chromosomes present, their small size, and their tendency to present themselves as pairs of parallel chromatides. However, HACKNEY (1974) succesfully applied chromosome counts in order to identify different species of *Meloidogyne*.

2.3. RELATIVE IMPORTANCE OF SPECIES OF *MELOIDOGYNE* AS INDICATED BY NUMBER OF LITERATURE REFERENCES

References from the world literature between 1949 and 1976 appearing in 'Helminthological Abstracts' were examined, and it was found that 93% of the references to identified species referred to the original species and subspecies described by CHITWOOD (1949) (See Table 1). Three of these species, *M. arenaria*, *M. incognita* and *M. javanica* are widely distributed through the tropics. In fact 76% of the references collected in Table 1 refer to *M. incognita*, *M. javanica*, *M. incognita acrita* and *M. arenaria*. These species are extremely polyphagous and constitute the main *Meloidogyne* problem in the tropics.

The other commonly cited species, *M. hapla*, inhabits cooler climates, though incidentally this species may be found in the tropics at high and cool altitudes (WITHEHEAD, 1969). The tropical species less frequently cited seem to be either restricted to one or a few hosts and/or to a limited geographical area. In this respect an exception should be made for *M. exigua* (Goeldi, 1887) Chitwood 1949, which has been found in Brazil, Central America and Trinidad and whose host range includes coffee (Coffea arabica L.), tea (Camellia sinensis (L.)

TABLE 1. Number of references to identified species of *Meloidogyne* reviewed in 'Helminthological Abstracts' between 1949 and 1976. Species marked by * are described by Chitwood in 1949.

Species	Number of references	Species	Number of references
*M. incognita**	778	*M. kikuyensis*	2
*M. javanica**	417	*M. litoralis*	2
*M. hapla**	300	*M. spartinae*	2
*M. incognita acrita**	159	*M. javanica bauruensis*	2
*M. arenaria**	93	*M. tadshikistanica*	2
M. naasi	49	*M. deconincki*	2
*M. exigua**	28	*M. africana*	1
M. graminicola	17	*M. phagosinae*	1
M. thamesi	15	*M. megadora*	1
M. graminis	9	*M. indica*	1
M. artiellia	8	*M. ethiopica*	1
M. coffeicola	7	*M. decalineata*	1
M. brevicauda	5	*M. lucknowica*	1
M. ovalis	3	*M. kirjanovae*	1
M. ottersoni	3	*M. oteifae*	1
M. inornata	3	*M. ardenensis*	1

Kuntze), pepper (Capsicum annuum L.) watermelon (Citrullus vulgaris Schrad.), and Bidens pilosa L.

Those species cited only once or a few times according to Table 1 seem not to represent a real threat to agriculture; however, they may be of great importance locally; for example *M. brevicauda* Loos, 1953, on tea in Ceylon. However, most species infrequently mentioned in world literature seem to be economically unimportant and the articles referring to these species are of taxonomic character. SASSER (1977) recently split the species of *Meloidogyne* into four groups: one consisting of species parasitizing grasses, one consisting of species mainly parasitizing trees and shrubs, one comprising species confined to limited areas (e.g. *M. litoralis*) and one consisting of polyphagous cosmopolitan species, frequently encountered (*M. incognita, M. javanica, M. arenaria* and *M. hapla*). Sasser estimated that the nematodes belonging to the last group account for more than 90% of the damage caused to economic crops.

2.4. THE SITUATION IN WEST AFRICA

Surveys made during the last fifteen years in West Africa have shown the frequent presence of the three most commonly occurring tropical species *M. incognita, M. javanica* and *M. arenaria* (ADDOH, 1974; CAVENESS, 1962; NETSCHER, 1970; NETSCHER & LUC, 1974). Since the suggestion of WHITEHEAD (1968) of examining females, juveniles from adhering egg-masses and males is not very practical, we attempted to develop a more simple procedure of identification which could possibly also be used by workers not necessarily nematologists and certainly not taxonomists.

In order to achieve this goal, we made observations and studies of several populations of *Meloidogyne* from West Africa; the results are presented below. This involved a series of published and unpublished studies dealing with improved techniques of fixation and mounting (NETSCHER & SEINHORST, 1969; NETSCHER, 1971; TAYLOR & NETSCHER, 1974), genetic studies concerning length of juveniles of species of *Meloidogyne* and *Heterodera* (NETSCHER & PERNES, 1971; NETSCHER, 1973), and studies on morphological variability of females of *Meloidogyne*. Because of the importance of the species *M. incognita, M. javanica* and *M. arenaria*, attempts were made to distinguish between these species only, thus neglecting all other existing species.

At first identifications were made on the basis of perineal pattern configurations, but the difficulties encountered by the workers already cited were also experienced. Another common problem was the very frequent appearance of natural populations composed of two or more species, an otherwise common phenomenon (OOSTENBRINK, 1957). Actually about 25% of the *Meloidogyne* populations present in the slide collection of the Laboratoire de Nématologie, ORSTOM, Dakar, Sénégal, are composed of more than one species.

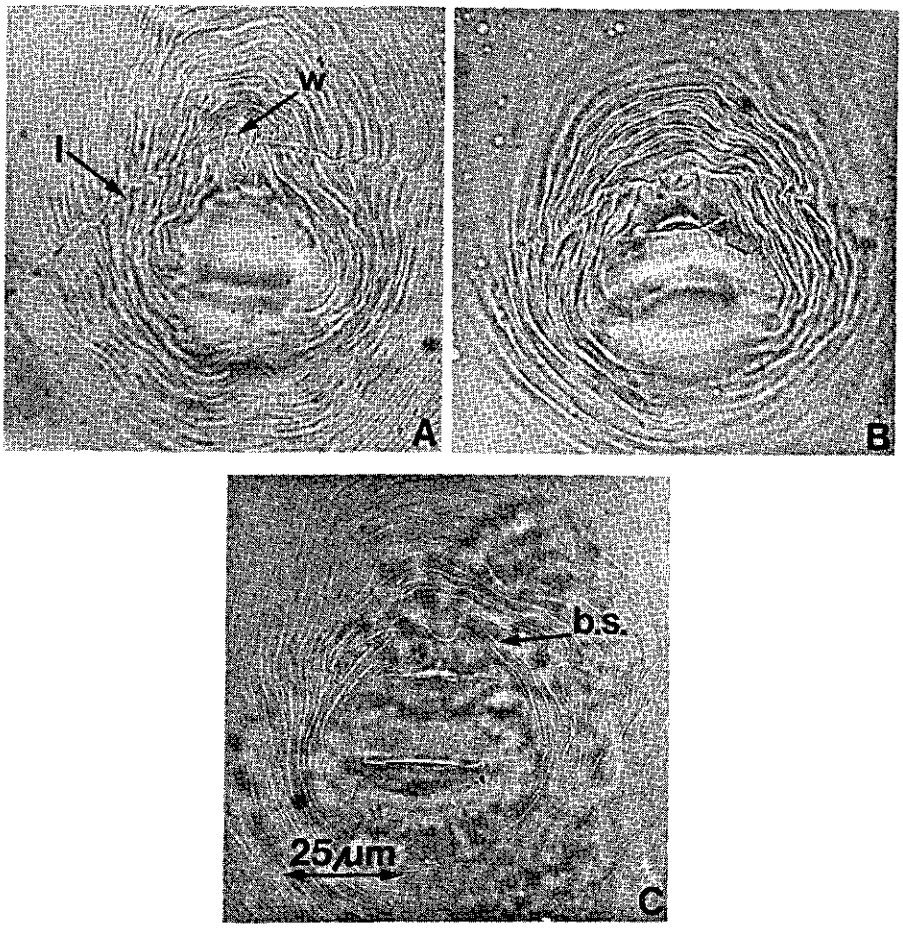

FIG. 1. Perineal patterns of females belonging to a wild population of *Meloidogyne* collected from egg plant (Population 3249 from Bambey, Senegal). A: *M. javanica*, l = lateral field, w = whorl; B: Perineal pattern intermediate between *M. javanica* and *M. arenaria*; C: *M. arenaria*, b.s. = striae broken at lateral field.

In the Senegalese populations particularly, we observed transitions from one species to another. To illustrate this, perineal patterns of a wild population collected from egg plant (Solanum melongena L.) at Bambey, Senegal, are shown in figure 1: fig. 1A closely resembles *M. javanica* as characterized by the lateral lines and the whorl in the tail-tip region; fig. 1C resembles *M. arenaria* (round pattern, with low dorsal arch, broken striae along lateral field). Fig. 1B possessed characters intermediate between the two. From the 27 females of this population examined, 9 were *M. javanica*, 9 *M. arenaria* and 9 were intermediate between these two species. Another population collected in the same garden on tomato (population 3251) consisted of the intermediate form only. In order to

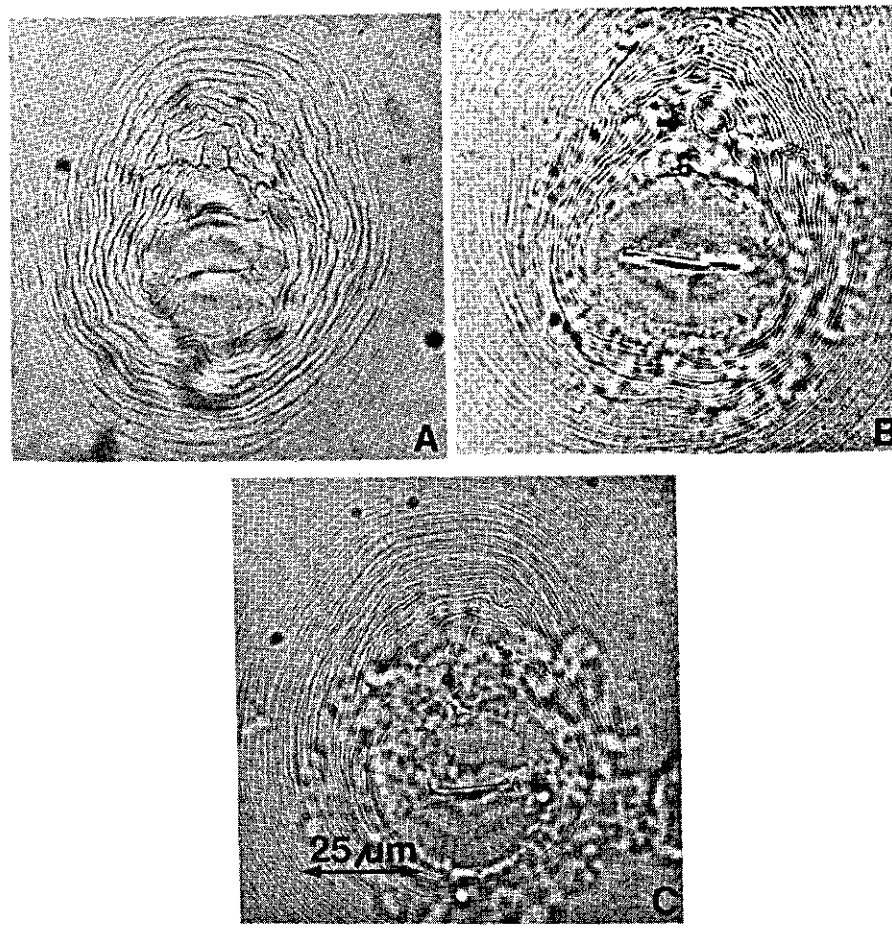

FIG. 2. A-C: Perineal patterns from a single egg-mass line derived from population 3249 with patterns predominantly attributable to *M. javanica*. (Compare variation in this pure line with that of wild population 3249 from fig. 1).

determine whether the intermediate patterns represented special forms, several single egg-mass cultures were established from the wild population and the patterns of these resultant lines were examined. Figs. 2A-C show a line with strong *M. javanica* tendencies and figs. 3A-C another with *M. arenaria* tendencies. Nevertheless in both lines intermediate types may be found (fig. 2C and 3C) that could belong to either of the two lines. In view of these observations. ALLEN's finding (1952) of females exhibiting *M. javanica* patterns in a pure line mainly composed of individuals of *M. incognita acrita* seems not too far fetched. Actually, Allen collected the *M. javanica* females from different hosts, including cotton (Gossypium hirsutum L.). To date, this plant has not been recorded as a host for *M. javanica* aside from a single report from MARTIN (1956); thus it

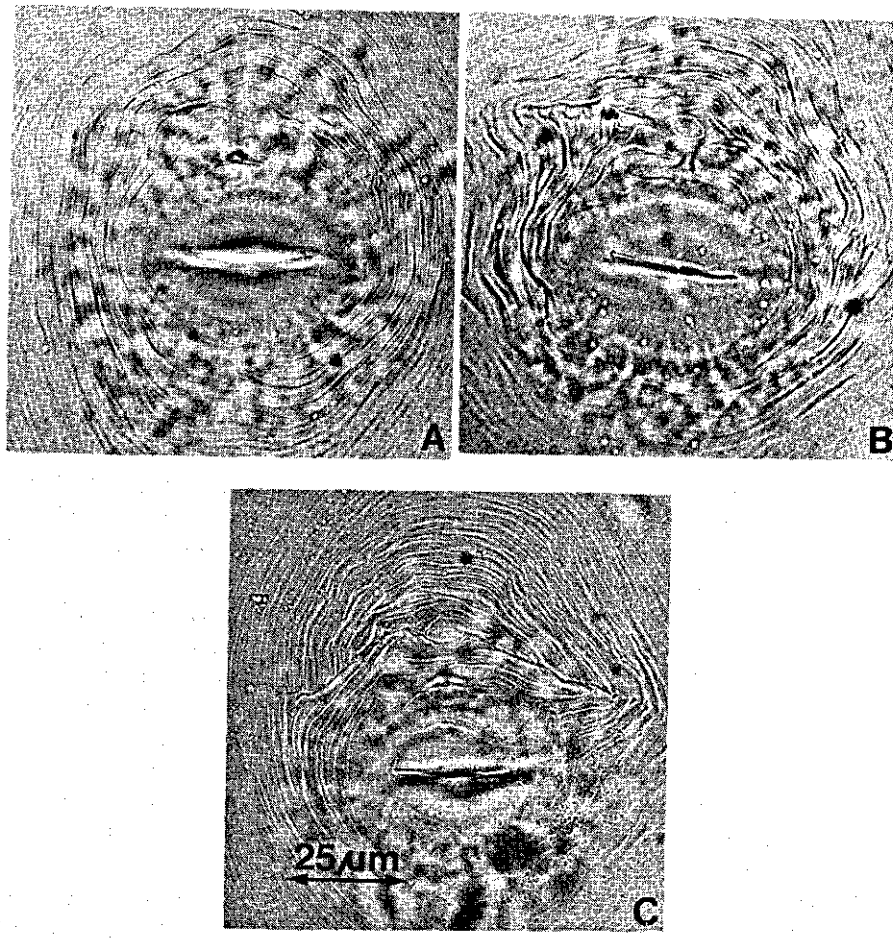

FIG. 3. A-C: Perineal patterns from a single egg-mass line derived from population 3249 with patterns predominantly attributable to *M. arenaria*. (Compare variation in this pure line with that of wild population 3249 from fig. 1).

seems unlikely that Allen's observations were caused by contamination of his *M. incognita acrita* line by *M. javanica*. We fully suscribe to his statement: 'It may be extremely difficult to determine if one is dealing with individual variation within a species or if two or more species are actually present' (ALLEN, 1952).

In addition to the intermediate forms discussed we found that single egg-mass lines of the *M. incognita* complex very often showed a variability in patterns within the ranges from *M. incognita* to *M. incognita acrita*, which is in agreement with the observations of TRIANTAPHYLLOU and SASSER (1960).

As the interpretation of perineal pattern morphology had proved to cause difficulties for the identification of field populations, measurements of the

TABLE 2. Vulval width of populations of *M. incognita* and *M. javanica* from West Africa. Each figure represents the mean, rounded to the nearest 0.5 μm, of 20 females measured.

M. incognita		*M. javanica*	
Population	Vulval width	Population	Vulval width
3278	20 μm	3264	21 μm
3432	20 μm	3205	22 μm
Adiopodoumé	21 μm	3444	23 μm
3298	21,5 μm	3458	24 μm
3270	23 μm	Ash 8	24,5 μm
3433	23 μm	3448	25,5 μm
M1 141	25 μm	3414	26,5 μm

vulval width of *M. incognita* and *M. javanica* were compared, as WHITEHEAD (1968) had shown that this character could be used successfully to separate these two species. Other measurements suggested by WHITEHEAD (1968) involving the exact position of the tailtip were rejected because of the difficulty of obtaining accurate data, since the precise position of the tailtip could not be definitely located in the majority of specimens examined.

Single egg-mass cultures of several *Meloidogyne* populations were established in tubes on root cultures of tomato using the technique described by NETSCHER (1973). From such cultures females were isolated, perineal patterns prepared and the vulval width measured. The means of 20 measurements made from 7 cultures of both *M. incognita* and *M. javanica* are presented in table 2. Results show that vulval width of females is not a reliable character to separate the two species. Statistical analysis agrees with this statement but shows that differences between populations are highly significant (table 3), suggesting that this character is under genetic control.

WHITEHEAD (1968) also determined the mean of the distance between phasmids of females and concluded that this character could be used to distinguish between *M. incognita* and *M. javanica*. According to these data (table 9, WHITEHEAD, 1968) the distance ranges from 25 to 28 μm in *M. javanica* compared to 21.1 μm for *M. incognita*. In the original description of *M. javanica*

TABLE 3. Analysis of variance of the data presented in table 2. ns: not significant, **: significant at 99% level.

Source of variation	Sum of squares	Degrees of freedom	Mean square	F
Meloidogyne species	179.20	1	179.20	3.03^{ns}
Populations	709.09	12	59.09	10.40^{**}
Error	1,509.7	266	5.68	
Total	2,397.99	279		

TABLE 4. Diagnostic characters of mature females of four tropical species of *Meloidogyne*, excluding characters of perineal patterns (after Esser, *et al.*, 1976).

Species	Stylet length	Position excretory pore to stylet knobs	Excretory pore*
M. incognita	15–16 μm	posterior	1/2
M. arenaria	14–16 μm	posterior	2
M. acrita	15–18 μm	–	–
M. javanica	14 (16) 18 μm	posterior	2½

* Position of excretory pore in relation to anterior end of female, expressed in stylet lengths.

(CHITWOOD, 1949) the interphasmidial distance of the type population was 19 μm, a much smaller figure than those given by Whitehead for either *M. javanica* and *M. incognita*. On the other hand in a single egg-mass population established from a population of *M. incognita* from Adiopodoumé, Ivory Coast, an interphasmidial distance of 26.5 μm was found, a value within Whitehead's range for *M. javanica*. On the basis of these figures, this character is rejected as a possible means of distinguishing between the two species.

ESSER, PERRY and TAYLOR (1976) recently proposed a series of female characters intended to distinguish between root-knot nematode species. Among these the configuration of the perineal pattern remained the most important character, but this time an attempt was made to describe the different parts of the pattern by coding the appearance of the striae by means of different letters. Excluding the characters describing perineal patterns, the remaining ones compiled for the West-African species of *Meloidogyne* are presented in table 4.

From table 4 it appears that the level of the excretory pore expressed in stylet lengths can be used to distinguish these species. Examining single egg-mass populations of *M. javanica* we found that this character ranged from 1.5 to 3.5, whereas in single egg-mass cultures of *M. incognita* and *M. incognita acrita* measurements were between 0.5 and 1.5. Taking measurements of *M. incognita* from figures 60a and c and 61 a presented by WHITEHEAD (1968) these data range between 1.5 and 1.8.

From the data presented here, its appears that *M. incognita* may be distinguished from *M. javanica* on the basis of the position of the excretory pore of the female, although a slight overlapping exists for this character between *M. incognita* and *M. javanica*. Thus to use this character for identification of *M. incognita* measurements of several individuals must be made and the results analyzed statistically in order to obtain reliable identification, assuming that future studies confirm these results. Values of this character for *M. arenaria* and *M. javanica* overlap, therefore these species cannot be distinguished from each other by making use of these measurements. The extreme variable position of the excretory pore within species and even pure lines of *Meloidogyne* again emphasizes the great morphological variability within the genus.

After examining numerous populations involving observations of thousands of females we did not find a method to identify every female of *Meloidogyne* with certainty. Though morphologically related groups certainly exist that fit the species concepts of CHITWOOD (1949), the great variability within pure lines and the abundance of mixed populations makes the identification of species of *Meloidogyne* based upon characters of females rather uncertain.

Frequently populations have been encountered which, based on perineal pattern morphology could have been considered as new species. However, experimentally determined host ranges always matched those of the other West-African populations. Cytological examination always revealed that the nematodes possessed many chromosomes (always over 40) and reproduced by mitotic parthenogenesis. On the basis of these facts we preferred to refer to such populations as *Meloidogyne* sp. instead of creating new taxa. The remarks of SANDERS and MULET (1976) who suggested that certain species of *Meloidogyne* might be morphological variants of existing species seem completely justified and the description of new species of *Meloidogyne* should be considered with caution. It seems advisable to include, if possible, cytological observations and host-range data in the description of new species of *Meloidogyne*.

WHITEHEAD (1968) and ESSER, PERRY and TAYLOR (1976) considered measurements and characters of juveniles of importance in determination of a species of *Meloidogyne*. The prospect of using juveniles to identify root-knot nematodes is highly tempting. Frequently juveniles of *Meloidogyne* are recovered from soil samples and specific determination in such cases involves inoculation of juveniles onto a suitable host in order to obtain mature females, necessary for the identification. Also it would be hoped that specific determination based on juveniles might confirm identifications based on females, especially in cases of ambiguity.

Total length was considered one of the most important characters of juveniles in species identification. To evaluate the validity of this character, preliminary studies were made with the amphimictic organism closest to *Meloidogyne* available, *Heterodera oryzae* Luc & Berdon, 1961. Studies of inbred lines of this nematode showed that with the techniques employed, influence of the environment on the variation of the length of juveniles ($\pm$ 50% of the variation) could be estimated. Further it was shown that within four generations of inbreeding the character under study, e.g. length of juveniles, was stabilized to a great extent (NETSCHER & PERNES, 1971). It was concluded that taxonomy based on quantitative characters should be based on the study of several populations. In fact taxonomy should be based on a number of characters both quantitative and qualitative.

Using the same techniques, a similar study of juvenile lengths of different single egg-mass populations of *M. incognita* and *M. javanica* from West Africa was made (NETSCHER, 1973). Comparison of the data obtained in West Africa and East Africa (WHITEHEAD, 1968) showed that this character, although suitable to distinguish between the East African populations, could not be used

for the West African populations because of overlapping that existed (table 5).

TABLE 5. Range of mean length of juveniles of *M. incognita* and *M. javanica* from populations obtained from East Africa and from West Africa.

East Africa		West Africa	
M. incognita	*M. javanica*	*M. incognita*	*M. javanica*
341–383 μm	391–426 μm	372–413 μm	388–439 μm

Further more it was shown that between the off-spring of sister females of *M. javanica* reared on the same root culture of tomatoes, mean length of juveniles differed significantly. Applying length as the only criterion for species identification, certain females of this line produced juveniles that should be identified as *M. incognita* whereas others should be considered *M. javanica*. These data are presented in table 6. On the basis of these results it was concluded that mean juvenile length could not be used to differentiate between *M. incognita* and *M. javanica* (NETSCHER, 1973).

TABLE 6. Mean length and standard deviation* of length of juveniles produced by three sister females of a pure line of *M. javanica* (after NETSCHER, 1973).

Mean length	Standard deviation
385 μm	1.7 μm
385 μm	2.8 μm
410 μm	1.5 μm

* Calculated for 15 juveniles.

The difference between juvenile length of *M. arenaria* and *M. javanica* reported by WHITEHEAD (1968) is debatable. WHITEHEAD (1968) and ESSER, PERRY and TAYLOR (1976) mention the same range of the juvenile length as given by Chitwood for the type specimens of *M. arenaria*: 450–490 μm; however, two populations of *M. javanica* with rather high mean juvenile length from Senegal (population 3414 and 3262 from table I in NETSCHER, 1973) had ranges

TABLE 7. Range of juvenile lengths of different populations of *M. javanica* and *M. arenaria*.

M. javanica		*M. arenaria*	
Population	Juvenile length	Population	Juvenile length
11C (CHITWOOD, 1949)	440–465 μm	Type population (CHITWOOD, 1949)	450–490 μm
3262 (NETSCHER, 1973)	415–461 μm	Race A (RIGGS & WINSTEAD, 1959)	364–535 μm
3414 (NETSCHER, 1973)	412–475 μm		

overlapping that of *M. arenaria*. The same was true for a population of *M. javanica* (11C) described in CHITWOOD's paper of 1949. These data are compared in table 7 together with the range of juvenile length of a population of *M. arenaria* studied by RIGGS & WINSTEAD (1959). On the basis of the evidence presented here it is not possible to distinguish between the most frequently occurring tropical root-knot nematodes, *M. incognita*, *M. javanica*, and *M. arenaria*, by means of the total length of juveniles.

ESSER, PERRY and TAYLOR (1976), quoting CHITWOOD (1949), distinguished *M. incognita* from *M. incognita acrita* by examining the rectum of juveniles of populations belonging to this group. They stated that *M. incognita* was characterized by a dilated rectum whereas in *M. incognita acrita* the rectum was not dilated. However, in a population of *M. incognita* from Abidjan, Ivory Coast, with perineal patterns having very pronounced 'acrita characters', every juvenile had a dilated rectum. Once again a discrepancy exists between observations of different workers due perhaps to the great variability existing in the genus.

The conflicting results obtained with juveniles may also be explained in part or totally by the different techniques used by different authors. Thus, CHITWOOD (1949) did not mention how specimens were killed, fixed and mounted; RIGGS & WINSTEAD (1959) killed juveniles by gentle heat and immediately measured them; Whitehead killed juveniles by gentle heat, fixed them in TAF, transferred them to hot lactophenol stained with cotton blue ans processed to glycerine by a rapid method (BAKER, 1953).

In our studies juveniles were obtained from egg-masses kept for 7 days in 0.3% mol Na Cl solution and subsequently transferred to water where hatching occurred during 3 days. Hatched juveniles were killed with hot FP 4:1 (NETSCHER & SEINHORST, 1969) fixed in 4% formalin and subsequently mounted in glycerine (SEINHORST, 1959). Juveniles of members of the Heteroderidae are liable to great changes in length (FENWICK & FRANKLIN, 1942) depending on the treatment they receive and possible measures should be taken in order to standardize techniques to obtain comparable data.

In West Africa, males of *Meloidogyne* were infrequently encountered in naturally occurring populations; thus, detailed studies of their morphology were not undertaken. When males were extracted from old greenhouse cultures, numerous intersexes and dwarf males were observed. Results of the detailed study of populations containing such aberrant forms were considered to be unreliable. A detailed study of the morphology of males might be very interesting from a taxonomic viewpoint; considering their infrequent occurrence in nature, however, male morphology in routine species identification is regarded to be of doubtful importance.

The validity of Terenteva's separation of *M. incognita* and *M. incognita acrita* on the basis of measurements of the height of the lip region of males of these species is doubtful (TERENTEVA, 1968). Only three populations were measured

(two of *M. incognita* and one of *M. incognita acrita*) and the ranges of this character slightly overlap for one population of *M. incognita* (5.8–6.9 μm) and the only population of *M. incognita acrita* (5.2–6 μm). Further it is difficult to attach much value to the precision of the measurements and the calculated means that are given to the tenth of μm. In our opinion Terenteva's figures should be rounded to 0.5 μm because this is the smallest unit of length that might be determined with confidence. If this is done, the mean hights of the lip region of males of the two populations cited should be 6 μm for both and not 5.7 and 6.2 μm. The small number of populations and the small number of individuals measured in Terenteva's study make the validity of her separation of the two species open to criticism.

The results of these studies may be summarized by stating that no infallibe methods have been found to identify naturally occurring root-knot nematode populations in West Africa to specific level. For every morphological character, there exists a vague area in which characteristics of one species overlap those of another.

3. HOST RANGE STUDIES

Before CHITWOOD (1949) reestablished the genus *Meloidogyne*, there were indications that different races existed within the root-knot nematodes, differing from one another in their host-parasite relations. Thus CHRISTIE and ALBIN (1944) investigating several populations from different regions of the U.S. found striking differences in comparative susceptibility of cotton, groundnut, and alfalfa to these populations. Groundnut was highly susceptible to three of the populations tested on this crop but highly resistant to the other populations: cotton was highly susceptible to one of the populations and resistant to the other populations tested; of the populations tested on alfalfa, 50% were capable of severe parasitism whereas this crop was fairly to highly resistant to the other populations.

After the description of the first species of *Meloidogyne*, inoculation trials of various plant species using pure lines of *Meloidogyne* could be utilized to establish host ranges for each of the known species. Knowledge of the host range of species of root-knot nematodes could be useful to develop crop rotations intended to reduce *Meloidogyne* populations, provided the populations could be identified to species. Unfortunately the host ranges of the three root-knot nematodes, *M. incognita*, *M. arenaria*, and *M. javanica* proved to be very extensive and included many crops. Table 8 indicates the economically important plant species which were listed as hosts of these three species (GOODEY *et al.*, 1965). Many economically important species belonging to many plant families, including Graminae, Leguminosae, Compositae, Solanaceae, Malvaceae and Cruciferae were included.

SASSER (1954) tested 50 plants against one population each of *M. incognita*, *M. incognita acrita*, *M. arenaria*, *M. javanica* and *M. hapla*. His host reactions are presented in table 9, modified by the synonymization of *M. incognita* and *M. incognita acrita* and the omission of 10 hosts which for some reason had not been tested to all five species of *Meloidogyne*. The following conclusions may be drawn from this table. Although in many cases all species of *Meloidogyne* react in the same way to a given plant, *M. hapla* clearly differs from the other species (in 13 of 40 cases). In the great majority of cases (34 out of 40) the three tropical species of root-knot nematodes reacted identically to a given plant: generally all three species were able to parasitize the plants tested, in only seven cases were the plants resistant. The reaction of four plant species, groundnut (Arachis hypogea L.), sweet pepper (Capsicum frutescens L.), cotton (Gossypium hirsutum L.) and sweet potato (Ipomoea batatas (L.) Lam.) appeared to be useful to differentiate between the tropical species of *Meloidogyne*. Using some of these species, Sasser suggested a series of differential hosts to facilitate identification of species of *Meloidogyne*. Later (SASSER, 1966) the number of host differentials was extended to include a greater number of plants.

TABLE 8. Partial list of economic plants, susceptible to *Meloidogyne incognita*, *M. javanica*, *M. arenaria* after GOODEY *et al.*, 1965.

Plant species	Common name	Family
Amaranthus caudatus L.	Inca wheat	Amaranthaceae
Amaranthus hybridus L.	Slim amaranth	Amaranthaceae
Amaranthus retroflexus L.	Redroot amaranth	Amaranthaceae
Celosia argentea L.		
Celosia cristata L.		
Carica papaya L.	Papaya	Caricaceae
Beta vulgaris L.	Beetroot	Chenopodiaceae
Beta vulgaris L.	Swiss chard	Chenopodiaceae
Beta vulgaris L.	Sugar beet	Chenopodiaceae
Helianthus annuus L.	Sunflower	Compositae
Lactuca sativa L.	Lettuce	Compositae
Brassica oleracea L. v. botrytis	Brocoli or Cauliflower	Cruciferae
Brassica oleracea L. v. capitata	Cabbage	Cruciferae
Brassica rapa L. spp. rapa	Turnip	Cruciferae
Raphanus sativa L.	Radish	Cruciferae
Citrullus vulgaris Schrad.	Watermelon	Cucurbitaceae
Cucumis melo L. v. reticulatus Naud	Melon	Cucurbitaceae
Cucumis sativus L.	Cucumber	Cucurbitaceae
Cucurbita maxima Duch.	Squash	Cucurbitaceae
Cucurbita pepo L.	Pumpkin	Cucurbitaceae
Ricinus communis L.	Castor bean	Euphorbiaceae
Avena sativa L.	Oats	Gramineae
Eleusine indica (L.) Gaertn.		Gramineae
Hordeum vulgare L.	Barley	Gramineae
Paspalum notatum Flugge	Bahia grass	Gramineae
Poa pratensis L.	Kentucky Blue grass	Gramineae
Saccharum officinarum L.	Sugar cane	Gramineae
Secale cereale L.	Rye	Gramineae
Triticum aestivum L.	Wheat	Gramineae
Zea mays L.	Maize	Gramineae
Glycine hispida Max. & G. soja Sieb. & Zucc.	Soybean	Leguminosae
Lupinus albus L.	White lupin	Leguminosae
Medicago sativa (L.) L.	Alfalfa	Leguminosae
Phaseolus vulgaris L.	Common bean	Leguminosae
Phaseolus mungo L.	Mung bean	Leguminosae
Pisum sativum L.	Pea	Leguminosae
Trifolium pratense L.	Red clover	Leguminosae
Trifolium repens L.	White clover	Leguminosae
Vicia faba L.	Broad bean	Leguminosae

TABLE 8. (continued)

Plant species	Common name	Family
Vigna catjung Walp.	Cowpea	Leguminosae
Allium cepa L.	Onion	Liliaceae
Hibiscus cannabinus L.	Kenaf	Malvaceae
Hibiscus esculentus L.	Okra	Malvaceae
Hibiscus sabdariffa L.	Roselle	Malvaceae
Ficus carica L.	Fig	Moraceae
Musa cavendishii Lambert	Cavendish banana	Musaceae
Passiflora edulis Sims	Passion fruit	Passifloraceae
Portulaca oleracea L.	Purslane	Portulacaceae
Prunus persica (L.) Zucc.	Peach	Rosaceae
Casimiroa edulis La Llave	White Zapote	Rutaceae
Lycopersicon esculentum Mill.	Tomato	Solanaceae
Solanum tuberosum L.	Potato	Solanaceae
Solanum melongena L.	Eggplant	Solanaceae
Nicotiana tabacum L.	Tobacco	Solanaceae
Camellia sinensis (L.) O. Kuntze	Tea	Theaceae
Daucus carota spp. sativus (Hoffm.) Thell.	Carrot	Umbelliferae
Pastinaca sativa L.	Parsnip	Umbelliferae
Vitis vinifera L.	Grape	Vitaceae
Zingiber officinale Rose	Ginger	Zingiberaceae

When more populations of different species of *Meloidogyne* were tested, differences in host reactions from those reported by SASSER (1954) were found. Although a complete literature review is not intended, a certain number of these exceptions are presented as example.

SASSER (1954) rated groundnut as host for *M. arenaria*, the so-called 'peanut root-knot nematode', and *M. hapla*. However, when several populations of *M. arenaria* were tested on groundnut variety 'florunner' (SASSER, 1966) only some of these populations reproduced on this host, whereas others did not. The same variability of reaction was obtained when 6 populations of *M. arenaria* from Florida were tested on groundnut (KIRBY *et al.*, 1975). SASSER (1954) listed groundnuts as a non-host of *M. javanica* and *M. incognita*. However, these observations have not always been confirmed by other workers. For example, groundnut has been attacked by *M. javanica* in Rhodesia (MARTIN, 1956),

Egypt (IBRAHIM and EL SAEDY, 1976) and Georgia (MINTON *et al.*, 1969): and OTEIFA, ELGINDI and MOUSSA (1970) and TAHA and YOUSIF (1976) mention groundnut as a host for *M. incognita* in Egypt.

SASSER (1966) found that cotton was severely attacked by 8 out of 18 populations of *M. incognita*; 4 populations did not attack cotton and the remaining six populations produced an intermediate reaction. KIRBY, DICKSON and SMART (1975) reported that only 4 out of 14 populations of *M. incognita* tested attacked cotton. SOUTHARDS and PRIEST (1976) studying 17 populations of *M. incognita* from Tennessee found striking differences in reaction towards cotton, pepper, cowpea (Vigna catjang Walp.) and watermelon (Citrullus vulgaris Schrad). They reported one isolate incapable of parasitizing watermelon and another not attacking pepper. Most isolates tested did not attack cotton and more than half of the populations did not reproduce on cowpea, a plant which we used for years at the Laboratoire de Nématologie of ORSTOM at Abidjan to rear *Meloidogyne* populations from West Africa including populations of *M. incognita*.

Strawberry (Fragaria ananassa Duch.) was originally reported as a non-host for *M. incognita*, *M. javanica* and *M. arenaria* (SASSER, 1954); this was supported by later work of SASSER (1966) and KIRBY, DICKSON and SMART (1975). NETSCHER (1970) tested the reactions of 25 populations consisting of either *M. javanica*, *M. incognita*, *M. arenaria* or a mixture of these species from Senegal to strawberry: none of the populations was able to parasitize this crop. In spite of this there are reports of attacks of strawberry by *M. javanica* or *M. incognita*. Thus MINZ (1958), MARTIN (1956) and TAYLOR and NETSCHER (1975) reported strawberry as a host for *M. javanica* in Israel, Rhodesia and Senegal respectively, while PERRY and ZEIKUS (1972) found a population of *M. incognita* that reproduced on strawberry.

Sweet pepper (Capsicum frutescens L.) is another differential host frequently utilized to distinguish between species of *Meloidogyne*. In the majority of cases *M. incognita* is able to parasitize pepper, whereas *M. javanica* does not (SASSER, 1966; KIRBY *et al.*, 1975). As previously mentioned SOUTHARDS and PRIEST (1976) reported that among 17 populations of *M. incognita* tested, one population did not reproduce on sweet pepper, the same result was obtained with two populations from Florida (PERRY and ZEIKUS, 1972). On the other hand, COLBRAN (1958) in Australia reported parasitism of pepper by *M. javanica*: however, he did not specify to which group of Capsicum frutescens the host belonged. A hot-pepper plant found in the field in Senegal was parasitized by both *M. incognita* and *M. javanica* (NETSCHER, 1970).

Additional cases of infraspecific variability in host parasite relationships are recorded in the literature and most probably, many new examples will be discovered. Nevertheless, it is believed that the evidence presented emphasizes that a physiological variability is no exception even within species of the genus *Meloidogyne*.

Additional proof of intraspecific variability within species of *Meloidogyne*

TABLE 9. Susceptibility of plant species to attack by root-knot nematodes (*Meloidogyne* spp.) adapted after SASSER (1954; table 1, p. 14 and 15).

Scientific plant name	Horticultural variety	Susceptibility rating*			
		M. incognita	*M. javanica*	*M. arenaria*	*M. hapla*
Allium cepa L.	Egyptian Winter	4	3	3	1
Amaranthus retroflexus L.		4	2	3	0
Ambrosia artemisiifolia L.		0	0	0	0
Arachis hypogaea L.	Spanish	0	0	4	4
Avena sativa L.	Arlington	1	1	1	0
Beta vulgaris L.	Detroit Dark Red	4	4	4	2
Brassica oleracea var. Capitata L.	Golden Acre	4	4	4	3
Calendula sp.		4	4	4	4
Capsicum frutescens L.	California Wonder	3	0	3	3
Citrullus vulgaris Schrad.	Dixie Queen	4	4	4	0
Crotalaria mucronata Desv.		0	0	0	0
Crotalaria spectabilis Roth.		0	0	0	0
Cucumis melo var. reticulatus Naud	Jumbo Hale's Best	4	4	4	1
Cucumis sativus L.	Marketer	4	4	4	1
Cururbita pepo L.	Early Prolific Straightneck	4	4	4	1
Daucus carota L.	Red Core Chantenay	4	4	4	2
Dianthus caryophyllus L.	Riveira Giant Mixed	3	3	2	1
(X Fragaria ananassa Duch.)	Blakemore	0	0	0	4
(X Fragaria ananassa Duch.)	Catskill	0	0	0	4
(X Fragaria ananassa Duch.)	Premier	0	0	0	4
(X Fragaria ananassa Duch.)	Temple	0	0	0	4
Glycine max (L.) Merr.	Hawkeye	4	4	3	4
Glycine max (L.) Merr.	Lincoln	3	3	3	4
Glycine max (L.) Merr.	Wabash	3	3	3	2
Gossypium hirsutum L.	Coker 100	3	0	0	0
Hibiscus cannabinus L.	PI. 189208c	4	4	4	0
Hibiscus esculentus L.	Clemson Spineless	4	3	1	0
Hordeum vulgare L.	Moore	4	2	2	0

TABLE 9. (continued).

Scientific plant name	Horticultural variety	Susceptibility rating*			
		M. incognita	*M. javanica*	*M. arenaria*	*M. hapla*
Impatiens sultanii Hook.		4	4	4	3
Impomoea batatas (L.) Lam.	Maryland Golden	2	0	0	2
Lycopersicon esculentum Mill.	Rutgers	4	4	4	4
Lycopersicon peruvianum (L.) Mill.		2	2	4	2
Medicago sativa L.	Atlantic	3	3	3	4
Nerium oleander L.		3	3	3	0
Nicotiana tabacum L.	Maryland Mammoth	4	4	4	3
Nicotiana tabacum L.	402	4	4	4	3
Pelargonium sp.	John Doyle	0	0	0	1
Pelargonium sp.	Rosana	0	0	0	1
Phaseolus vulgaris L.	State Half Runner	3	4	4	4
Phaseolus vulgaris L.	Top Crop	4	4	4	4
Pisum sativum L.	Thomas Laxton	4	3	4	3
Raphanus sativus L.	Early Scarlet Globe	4	4	4	3
Rhododendron sp.	Glenn Dale Hybrid	0	0	0	0
Secale cereale L.	Prolific	3	2	2	0
Solanum melongena L.	Black Beauty	4	4	3	4
Solanum tuberosum L.	Irish Cobbler	4	3	3	3
Strophanthus sarmentosus D.C.		1	0	0	0
Triticum aestivum L.	Coastal	4	4	3	0
Zea mays L.	Cogent Ill. 8 × 6	3	3	3	0
Zea mays L.	Golden Cross Bantam	3	3	2	0

* 0: no infection or if larvae entered the roots they did not develop into mature egg-laying females; 1: extremely light infection with only an occasional mature female with egg-mass; 2: light infection with mature females and egg-masses easily seen with the naked eye; 3: moderate infection with full grown females and egg-masses moderately abundant; 4: severe infection with mature females and egg-masses very abundant.

is obtained by studying varieties of normally susceptible plant species that have been bred for resistance to species of *Meloidogyne*. Among susceptible crops resistant cultivars have been developed for beans (Phaseolus vulgaris L.), cowpeas, soybeans (Glycine hispida Max. & G. soja Sieb. & Zucc.), sweet potatoes, and tomato (Lycopersicon esculentum Mill.). As resistant varieties are selected by testing the off-spring of crosses of resistant × susceptible parents to one or a few *Meloidogyne* populations only, it is not surprising that populations exist that react differently when these varieties are tested to many populations. For example, the sweet potato varieties Allgold and Puerto Rico were reported to be resistant to *M. arenaria* (GIAMALVA *et al.*, 1963). Nevertheless the reactions of these varieties, when tested against different populations of this nematode, ranged from completely resistant to highly susceptible (SASSER, 1966; KIRBY *et al.*, 1975). Another resistant variety used as a differential host (SASSER, 1966) is the tobacco variety N.C. 95 which is resistant to *M. incognita*. Nevertheless, of 32 populations of *M. incognita* tested (SASSER, 1966; KIRBY *et al.*, 1975), four successfully reproduced on this variety. In addition a naturally occurring race of *M. incognita* from the eastern shore of Virginia reproduced well on the 'resistant' tobacco variety N.C. 95 (PHIPPS *et al.*, 1972).

Many studies have been made on tomato varieties resistant to *Meloidogyne*. RIGGS and WINSTEAD (1959) reported that one population each of *M. incognita*, *M. incognita acrita* and *M. arenaria* reproduced only slightly on resistant tomato varieties. By successive transfers of the juveniles from egg masses produced on these resistant tomatoes to other resistant tomato plants, populations were developed capable of breaking this varietal resistance. They termed these populations 'B races'.

This phenomenon was also observed by TRIANTAPHYLLOU and SASSER (1960), who reported that certain clones of *M. incognita* they studied (either cultures derived from single egg masses or single juveniles) were able to develop 'B races' on resistant tomato. ALLEN (1952), studying 6 different populations obtained from cotton fields, probably all *M. incognita*, reported that two were able to reproduce slightly on Lycopersicon peruvianum (L.) Mill., the species from which *Meloidogyne*-resistance in tomato, Lycopersicon esculentum Mill., was derived. NETSCHER (1977) stated that certain populations of *M. javanica* and *M. incognita* from West Africa were able to develop 'B races', whereas others did not. Indications were obtained that the process of adaptation to resistant varieties was gradual. Further it was observed that certain naturally occurring populations of *M. incognita* and *M. javanica* were capable of severely attacking resistant tomato varieties.

SIDHU and WEBSTER (1974, 1975) showed that resistance to *M. incognita* in tomato was determined by two dominant genes $LMiR_1$ and $LMiR_2$ which were closely linked and that $LMiR_1$ and Mi, the gene that determines the resistance of the tomato variety Anahu are either allelic or identical. During a study of populations from Senegal, one of *M. incognita* (Nr. 11304) and one of *M. javanica* (Nr. 11310) were found to reproduce on the resistant variety, Rossol.

Both populations were found capable of reproduction on the variety Nematex, possessing the gene $LMiR_1$ and Small Fry possessing $LMiR_2$. Another isolate of *M. incognita* (Nr. 11575) reproduced on Small Fry, but did not reproduce on Rossol and Nematex. These data are presented in table 10. On the basis of these data it is suggested that a gene for gene relationship exists in this case between genes for resistance in tomato, and for virulence in *Meloidogyne*. Populations of *Meloidogyne* not attacking any resistant varieties should be characterized by 'o', populations attacking varieties with $LMiR_1$, by '1', populations capable of parasitizing varieties with $LMiR_2$ by '2', and populations reproducing on varieties possessing the two genes $LMiR_1$ and $LMiR_2$ by '1, 2'. (See table 10).

For the moment 3 physiological races have been identified in Senegal, one of *M. javanica* ('B race' Nr. 11310, Type '1, 2') and two of *M. incognita* ('B race' Nr. 11304, Type '1, 2' and Nr. 11575, Type '2'). Future studies of additional *Meloidogyne* populations are necessary to characterize the populations as far as virulence towards resistant tomato varieties possessing the genes $LMiR_1$ and/or $LMiR_2$ is concerned.

TABLE 10. Reaction of populations of *Meloidogyne* to resistant tomato varieties.

			Variety and resistance genes			
Species	Population	Type	Roma (universally susceptible) $LMir_1$ $LMir_2$	Nematex $LMiR_1$	Small Fry $LMiR_2$	Rossol*
M. javanica M. incognita	11310 11304	1, 2 1, 2	+	+	+	+
M. incognita	11575	2	+	−	+	−
To be found	–	1	+	+	−	−
M. javanica M. incognita	Majority of populations	0	+	−	−	−

* As it is not known whether Mi and $LMiR_1$ are identical or allelic, and since the reactions observed make it not possible to deduce whether Rossol possesses $LMiR_2$, the following gene combinations are proposed for this variety:
$LMiR_1LMiR_2$, $LMiR_1$ $LMir_2$, $MiLMiR_2$, and $MiLMir_2$.

The data presented here demonstrate that the tropical species of *Meloidogyne* even in a geographically limited area such as Senegal exhibit strong physiological diversity as measured by variability in host-parasite relationships. Theoretically amphimictic species should have an advantage over parthenogenetic species as the processes of meiosis and fertilization assure the amphimictic organism of a wealth of different genotypes whereas reproduction of mitotic parthenogenetic species essentially results in the establishment of clones. Therefore the parthenogenetic nature of the species of *Meloidogyne* considered here is difficult to reconcile with the diversity observed in these organisms. However, in certain cases, parthenogenetic reproduction may be more favourable for the establishment of certain specialized populations. As TOXOPEUS (1956) pointed out, adapted races to resistant varieties whose creation is based upon a mutation from a dominant to a recessive gene are difficult to realize in amphimictic species, as it involves the mating of females and males both possessing the rare recessive gene. Mutation in a parthenogenetic species automatically results in the creation of a new clone characterized by the mutant gene. If a mutation in a parthenogenetic population provides the mutated individual with the potential capacity to parasitize a resistant variety, a 'B race' will be automatically selected when it happens to parasitize a resistant plant.

Our findings that of a number of populations tested on a resistant variety of tomato only a few were able to develop 'B races' and that the development of the agressive populations was rather a gradual than an abrupt change (NETSCHER, 1977) are not in agreement with the hypothesis that the creation of 'B races' was caused by a single mutational step. It rather seems that a selection among increasingly agressive individuals in the populations was accomplished by the resistant variety. Different hypotheses may be forwarded to explain this gradual adaptation.

It may be possible that subsequent mutations from low to high agressivity of the virulent race took place on corresponding loci of homologous chromosomes of the polyploid population of *Meloidogyne* concerned, resulting in the establishment of the 'B race'. As a supplementary hypothesis one must suppose that the effect of each mutation is additive.

Another hypothesis intended to explain the adaptation of *Meloidogyne* populations to resistant varieties might be that cytoplasmic heterogeneity of the ova is responsible for the genetic diversity within pure lines of *Meloidogyne*. To explain certain adaptive processes in the saprozoic nematode *Caenorhabditis elegans* it was assumed that a certain degree of cytoplasmic heterogeneity was present in this organism.*

* BRUN (1966) observed that normally reared at 18°C, *Caenorhabditis elegans* Maupas, 1900, could be gradually adapted to 24.5°C by transferring the animals for many generations at intermediate temperatures; when after a sufficient number of generations the nematodes were adapted to an intermediate temperature, they were transferred to a higher temperature and this process was repeated till the temperature of 24.5°C was reached. His observations led him to suggest that there was 'a progressive production of adapted and transmittable cytoplasmic states which would be responsible for the begetting of lineages fertile at high

Whatever the process may be that causes heterogeneity of the mitotic parthenogenetic species of *Meloidogyne*, it will be difficult to elucidate as no methods have been developed to study the genetics of animals not reproducing sexually. Therefore, the hypotheses presented are all speculative.

Much additional information must be provided before the factors determining host-parasite relationships between plants and nematodes will be completely known. In the Heteroderidae the crucial point is whether or not syncytia ('giant cells') are formed, once the nematodes have penetrated a given plant. According to BIRD (1974), 'It seems that if *Meloidogyne* species are to reproduce normally, they must be able to initiate and maintain these syncytia'. In a review article on plant response to root-knot nematodes (BIRD, 1974), a highly complex picture was developed of the different physiological and biochemical relationships between the stimuli from the nematode and the responses of the host. The author emphasized that the nematode interferes with the differentiation of the parasitized cells and stated: 'During syncytial formation the nematode induces specialized cells to be formed from unspecialized cells. These unspecialized cells would normally form specialized cells of different types with specific functions and response to various repressors and inducers operating within the plant. The nematode also represses part of the cell's genetic coding and activates others so that a special type of cell is induced'.

It seems logical that the polyphagous species of root-knot nematodes induce the same processes in the different host plants which they parasitize and that the nematodes interfere in a group of functions that these plants have in common. In such a complex relationship relatively minor genetic differences in the host may upset the delicate balance between stimulus of the nematode and response of the host resulting in the malfunctioning or repression of syncytia and hence resistance of the host. The fact that several cases of resistance to root-knot are known in susceptible crops that depend on only one or a few genes (KEHR, 1966) support this point of view. Similar minor differences in the genotype of the nematode may account for the existence of populations capable of parasitizing resistant plants.

Different types of resistance to root-knot nematodes are known. The most commonly reported reaction of host tissues is hypersensitivity resulting in necrosis of the cells in the vicinity of the nematode (RIGGS & WINSTEAD, 1959: GIAMALVA *et al.*, 1963: MINTON, 1963: DROPKIN, 1969: NETSCHER, 1975). GIEBEL (1974) proposed a model, largely experimentally supported, to explain

temperatures'. Although the gradual adaptation of *Meloidogyne* clones to resistant varieties may show points of similarity with the situation described by Brun for the adaptation of *C. elegans* to high temperatures, the selection of resistance breaking forms of *Meloidogyne* is too rapid to be completely comparable with the situation of *C. elegans* where for each step of the adaptation hundreds of generations were required. Brun quoting HINSHELWOOD (1947) remarked that from a physiological point of view, the adaptation to temperature is a process necessitating more profound changes in the cellular material than those that accompany the adaptation to drugs or new substrates.

the susceptibility/resistance mechanism of varieties of potato (Solanum tuberosum) resistant towards *Heterodera rostochiensis* WOLLENWEBER, 1923. Glycosidases and proteases released by the nematodes on the plant's phenolic glycosides, conjugate auxins, and proteins induce a number of interdependant reactions resulting in a resistant or a susceptible response, depending on the chemical composition of the cellular constituents of the plant varieties and those of the substances released by the nematode. Although species of *Meloidogyne* generally provoke galls and are more polyphagous than species of *Heterodera*, the necrotic hypersensitive reaction of plants resistant to *Meloidogyne* closely resembles the response of resistant potato varieties to invasion by *H. rostochiensis* and could be due to analogous processes.

In addition to the necrotic reaction to invasion by juveniles of *Meloidogyne*, other reactions have been reported which protect the plant from the parasites. GIAMALVA, MARTIN and HERNANDEZ (1963) suggested that apart from the necrotic reaction, sweet potato varieties resistant to species of *Meloidogyne* possessed another mechanism of resistance which was more favourable to yield than hypersensitivity which provoked intensive necrosis of roots and tubers. JATALA and RUSSELL (1974) studying the nature of sweet potato resistance to *M. incognita* suggested that resistance may be based in part on the synthesis of nematode-repellant exudates and failure of the juveniles to penetrate the plant. In most cases in which *Meloidogyne* resistant plants were exposed to the nematodes, detailed observations have shown that juveniles did penetrate the plants.

Thus, BARRONS (1939) could not show significant difference in the number of juveniles observed in 19 resistant plant species belonging to 14 genera and five good hosts of root-knot nematodes. DROPKIN (1969) studying the effect of temperature on the necrotic reaction of resistant tomatoes and other poor hosts of *Meloidogyne incognita acrita* reported that juveniles penetrated all plants tested. The fate of the nematode depended on the reaction of the plant which was influenced by temperature in the majority of cases.

A recent survey of weeds commonly associated with vegetable crops in Senegal has shown that these plants were penetrated by juveniles when inoculated with *M. incognita* or *M. javanica* regardless of their host-status (HUOT, pers. comm.).

When Lantana sp. was exposed to juveniles of *Meloidogyne* sp., virtually no penetration was observed (CHRISTIE, 1949); the same observation was made on Tagetes patula L. when this plant was inoculated with either *M. javanica*, *M. arenaria* or *M. incognita* (WINOTO, 1969). Although these observations suggest that these plants are not readily penetrated by juveniles of *Meloidogyne* it is no proof. Sometimes juveniles of *Meloidogyne* that have penetrated an unsuitable host are capable of leaving such a plant (DE GUIRAN, 1961; REYNOLDS *et al.*, 1970). Penetration by *Meloidogyne* may also be overlooked if the period between inoculation and observation is too long, especially when a necrotic reaction is involved causing the attacked roots to decompose.

On the basis of the observations presented, it seems likely that most plant

species are attacked by *Meloidogyne*, although exceptions may exist. Observations by MERNY and CADET (in press), who observed that juveniles of *Heterodera oryzae*, a parasite restricted to rice (Oryza sativa L.) and maize (Zea mays L.) were capable of penetrating plants such as tomato and soybean, emphasize the potential tendency towards polyphagy within the Heteroderidae. In summary, it can be stated that penetration of a plant by juveniles of *Meloidogyne* and other members of the Heteroderidae is no indication of its host status. As a consequence, it seems reasonable that in rare cases a *Meloidogyne* individual possessing a slightly altered genotype may enter a 'non-host', and because of its modified physiology may be able to establish a successful host-parasite relationship giving rise to a new physiological race. Thus, numerous examples exist of populations of species of *Meloidogyne* reproducing on 'non-hosts' (as reported in the literature), and populations of these species ('B races', 'biological races') are frequently encountered. The reason we prefer to emphasize exceptions rather than the rule is to stress that the *Meloidogyne* situation is much more complex than much of the literature suggests. Although other workers have reported *Meloidogyne* populations that do not confirm to published host-lists, the importance of these reports is generally overlooked in an attempt to simplify the complicated problem.

This physiological variability together with the morphological variability discussed in the preceding section tend to support the point of view of TAYLOR (1976) who suggested that, 'From the practical point of view, one must ask whether or not we are much further ahead today in understanding the *Meloidogyne* problem than when this group of pathogens was referred to as '*Heterodera marioni*' with several recognized physiological races'.

4. THEORETICAL AND PRACTICAL IMPLICATIONS OF THE VARIABILITY OF *MELOIDOGYNE*

From the preceding sections it is evident that as far as morphology and host preference are concerned, great intraspecific variability exists within the species *M. incognita*, *M. javanica* and *M. arenaria*, and that for all characters utilized in specific determination, overlapping exists between species. From a taxonomic point of view this situation is difficult to accept, and it is not surprising that more or less contradicting opinions exist. Thus, TRANTAPHYLLOU (1962) stated: 'Most root-knot nematodes may reproduce exclusively by parthenogenesis and, therefore, their classification into species is not possible, since the species concept does not apply to such organisms', whereas FRANKLIN (1971) considered the species concept in *Meloidogyne* valid. She stated: 'The concept of reproductive isolation is irrelevant in parthenogenetic populations such as most *Meloidogyne* species. But *Meloidogyne* populations distinctive enough both morphologically and biologically to be recognized as species are well known'. Referring to SIMPSON's (1961) definition of 'evolutionary species', which envelops both biparental and uniparental populations, she accepted the validity of the parthenogenetic species of *Meloidogyne*.

An example of differences of opinion concerning validity of taxa within *Meloidogyne* is as follows: CHITWOOD (1949) when reestablishing the genus *Meloidogyne* described a population as the variety *M. incognita acrita* which later was considered as a subspecies. The acceptance of *M. incognita acrita* as a taxon has led to a certain controversy. TRIANTAPHYLLOU and SASSER (1960) synonymized *M. incognita* and *M. incognita acrita*, whereas GOLDEN (1974) distinguished between at least three different forms in the *M. incognita* group and later GOLDEN (1976) considered *M. incognita* as a group consisting of four recognizable subspecies plus one or more races. ESSER, PERRY and TAYLOR (1976) recently elevated *M. incognita acrita* to specific rank by creating *M. acrita* (Chitwood, 1949) Esser, Perry and Taylor, 1976.

Exactly the opposite position was taken by USTINOV (1959) who did not accept the division of *Heterodera marioni* into separate species of *Meloidogyne* and proposed the binomial *Meloidogyne marioni* to include all species of the genus known at that date.

USTINOV's (1959) rather intuitive approach was based mainly upon reports from literature. Since then a wealth of studies have been published including the cytological observations of TRIANTAPHYLLOU (1962, 1964, 1966, 1971) that made it possible to show the phylogenetic relationships between species of *Meloidogyne*. In view of knowledge acquired since 1959, it is desirable to reconsider the taxonomic position of the most abundant and most studied species of *Meloidogyne*, especially in view of the possible practical implications to agriculture. Instead of trying to separate species from each other on the basis of characters, known to be highly variable, it is proposed to group populations

on the basis of characters they have in common. Thus a group including *M. incognita*, *M. javanica* and *M. arenaria* can be separated from other species described to date. These three species are characterized by their wide distribution and polyphagous nature, their mitotic parthenogenetic mode of reproduction and the great number of hosts they have in common (see tables 8 and 9). DALMASSO and BERGÉ (1975), discussing the genetic variability of species of *Meloidogyne*, mentioned the 'complexe mitotique' consisting of *M. incognita*, *M. javanica*, and *M. arenaria*, a concept to which we fully suscribe, as it is in agreement with the data presented in Sections 2 and 3.

Although the acceptance of '*Meloidogyne marioni*' as a species, grouping the tropical 'mitotic complex', is tempting, it is not possible to do so as this designation was reserved as a possible synonym of *M. hapla* (Chitwood, 1949; Chitwood in CHITWOOD, SPECHT & HAVIS, 1952); and BAKER (1962) considered *M. marioni* (Cornu, 1887) Chitwood & Oteifa, 1952, a *species inquirenda*. The acceptance of *M. marioni* as a valid species also would complicate the taxonomic position of the described species *M. incognita*, *M. javanica* and *M. arenaria*, since they would then be considered at the level of subspecies. Therefore, we prefer to consider those tropical polyphagous populations of *Meloidogyne* characterized by mitotic parthenogenesis as a group. This group includes the described species *M. incognita*, *M. javanica*, and *M. arenaria*, populations possessing characters intermediate between these species, possibly undescribed species, and perhaps certain described species whose physiological and cytological properties have not yet been investigated (a species like *M. ethiopica* Whitehead, 1968, could be part of this complex). When the word 'group' is proposed, it is done deliberately to indicate the great resemblance between members of populations sharing similar characters. However, in certain cases, sufficient differences exist to separate populations belonging to one species from another species. Thus, for example, when a nematologist cooperates with a plant breeder in developing a *Meloidogyne* resistant cotton variety, he will provide the breeder with different populations of *M. incognita* that are capable of parasitizing cotton, while ignoring *M. javanica* populations (unless he possesses populations of the latter species which attack cotton). We believe that by recognizing the existance of the complex, a greater emphasis is placed on the characters which the tropical species have in common and that the specific determination as is practiced now (mainly based on the configuration of the perineal pattern) does not really serve much purpose. In certain cases, attempts to identify species may even lead to incorrect decisions. The following example will illustrate this point of view.

In 1974, a rotation experiment was carried out at the Centre de Développement d'Horticulture at Cambérène, Senegal, intended to reduce the population level of root-knot nematodes, composed of a mixture of *M. incognita*, *M. javanica*, *M. arenaria*, and intermediate forms (determined according to perineal patterns). On the basis of host-range studies of SASSER (1966) and KIRBY, DICKSON and SMART (1975) the following predictions should have been made:

strawberry should not be parasitized by the *Meloidogyne* population present (because of the absence of *M. hapla*), whereas groundnut might well be parasitized by the *M. arenaria* population. However, the following actually occurred: groundnut was not parasitized by the root-knot nematodes and populations dropped considerably under the influence of this crop (NETSCHER, 1974). The same population contained nematodes capable of parasitizing strawberry and within 6 months a population of *M. javanica* developed on this host (TAYLOR & NETSCHER, 1975). Thus, if the choice of the non-host in the rotation had been based on specific determination (perineal patterns) and published host-range data, strawberry would have been chosen as a rotational crop to reduce the *Meloidogyne* population and groundnut would have been avoided. On the basis of results obtained, exactly the contrary should have been recommended.

The example cited illustrates the danger of total reliance on published host-ranges for certain species of *Meloidogyne* without studying the actual physiological variation of the populations concerned. One might even question the usefulness of making specific determinations intended for recommendation of crop rotations. Although it may be of interest to a nematologist to know that the Chambérène populations harbour a race of *M. javanica* capable of parasitizing strawberry, farmers working at Cambérène are not interested in knowing whether it is 'species A' or 'species B'. What is important to them is that a root-knot problem exists on strawberry in their fields. Testing this particular population without previous identification of the 105 females that were used in this study would not have changed the results.

In most countries where tropical root-knot nematodes are a problem, there are very few nematologists (in a vast country like Indonesia, *Meloidogyne* is being investigated by one person only) and the expertise of these workers should be employed in the most efficient manner. Therefore, instead of identifying root-knot nematodes, a highly specialized technique requiring an experienced nematologist, tests with local populations of *Meloidogyne* on crops of potential interest to the farmers should be made by agronomists, extension officers, etc. in cooperation with a nematologist. Crops to be tested should include cash crops, cover crops, food crops and resistant varieties of susceptible crops.

Another danger of relying on the identification of field populations to species is that sometimes one of the species belonging to the population is so rare that it will not be detected. Thus, by growing groundnut in a field thought to be infested only by *M. javanica*, SAUER and GILES (1959) increased a population of *M. hapla* that had escaped their attention. In a similar way a population of *M. incognita* collected on tobacco in Senegal (population 11317) did not parasitize sweet pepper, apart from two females that proved to be *M. arenaria*. Their off-spring readily reproduced on sweet pepper (NETSCHER, unpublished). In order to detect these rare individuals, nematode populations to be tested should be sufficiently large, and field tests are preferred to glasshouse tests.

Because of the great physiological variability of root-knot nematodes, the host-ranges of a large number of populations should be determined in each

area (ideally populations from each infested field should be tested), something feasible in countries with skilled farmers and well organized extension services. In countries where these requirements cannot be met, and a limited number of populations has been examined, a more statistical approach should be taken. Of 25 single egg-mass cultures obtained from the same number of field populations from Florida, KIRBY, DICKSON and SMART (1975) found that only two reproduced on strawberry and six on groundnut. It is mathematically logical to select these two crops in Florida, if one wishes to decrease root-knot populations by crop rotation. Use of single egg-mass cultures should be avoided for this kind of study, because such cultures do not represent the physiological variability of the original populations from which they have been derived. From field observations and studies carried out in Senegal it may be concluded that groundnut is rarely if ever parasitized (NETSCHER, 1975), strawberry seldom (NETSCHER, 1970; TAYLOR and NETSCHER, 1975) whereas resistant tomato varieties are fairly often parasitized (NETSCHER, 1977).

In breeding for resistance to *Meloidogyne*, physiological variability of different populations of root-knot nematodes must be kept in mind. Most root-knot resistant varieties have been developed by testing the progeny of crosses against one or only few *Meloidogyne* populations. Thus resistance in certain breeding lines or related species may have been overlooked. This is well illustrated by the work of MARTIN and BIRCHFIELD (1973), who found that the sweet potato variety 'Centennial' susceptible to *M. incognita*, showed a strong hypersensitivity reaction when it was inoculated with a population of *M. incognita* isolated from soybean: on the other hand, the resistant sweet potato variety 'La 4-73' was heavily attacked by a population of this nematode in Maryland. Two additional hypothetical examples, based upon data reported in the literature, will provide a better understanding of the complications involved in breeding varieties resistant to an organism as variable as the root-knot nematode. The 'Albizzia race' of *M. incognita* found on the Eastern shore of Virginia readily parasitizes the resistant tobacco variety 'NC 95' (PHIPPS *et al.*, 1972); therefore, this variety would never have been developed if screening for resistance had been done in a field containing this race. In the same way, the resistant tomato varieties developed in the United States carrying the genes Mi (or $LMiR_1$) and $LMiR_2$ would not have been recognized, had they been tested in areas of Senegal where naturally occurring resistance-breaking biotypes occur (NETSCHER, 1977). Therefore, it is recommended that all sources of resistance to *Meloidogyne* should be tested to many populations occurring in the world. Thus certain germplasm may be susceptible in a certain area but may be resistant in another. Conversely it may be found that certain sources of resistance will be susceptible in other areas. A mechanism such as the International *Meloidogyne* Project, organized by the State University of North Carolina, and sponsored by U.S. A.I.D. could be of great value in the exchange of germplasm between countries as well as to provide useful information to and from nematologists working throughout the world.

On the basis of considerable experience in the tropics studying *Meloidogyne* problems, the use of non-hosts and resistant varieties should be recommended on slightly infested or noninfested land:

1. In most cases reduction of the nematode population is accomplished by the trapping effect of the roots of the plants involved, accompanied by necrosis of the invaded root tissues. If the *Meloidogyne* populations are too large, plants are badly damaged and a serious weed population, generally comprising several hosts of *Meloidogyne*, will develop with a resultant multiplication of the root-knot nematodes. In one extreme case, we observed a complete failure of a crop of groundnut grown with the intention of reducing the root-knot population in a heavily infested field; among the weeds that had become dominant to the groundnuts, many were actively maintaining the *Meloidogyne* population, based upon the degree of galling of the roots. From inoculation experiments, it was shown that population densities of 32,000 juveniles per dm^3 of soil or more, a level frequently encountered in the field, severely damaged groundnut seedlings.
2. It is also important to avoid growing non-hosts in heavily infested soils so as to reduce the possibility of selecting and developing biological races from the existant *Meloidogyne* populations. Assuming that the frequency of such biological races is independant of the number of the nematodes present, it is logical to assume that in soils containing few root-knot nematodes, these races will be rare or absent.

For these two important reasons it has been recommended to use crop rotation and resistant varieties primarily as preventive measures rather than as a cure for the *Meloidogyne* problem (NETSCHER, 1974, 1975, 1977; NETSCHER and MAUBOUSSIN, 1973). In cases of heavy infestations, *Meloidogyne* populations should be reduced by either chemical or other means (soil desiccation, inundation, bare fallow) before growing a non-host or a resistant variety.

Under ecological conditions occurring commonly in Senegal, root-knot nematodes do not represent a great danger to susceptible crops. In Senegal, cowpea was very susceptible to all root-knot nematode populations tested, and yet under field conditions no extensive damage due to *Meloidogyne* has been reported. The Senegalese wet season lasts about four months a year, just long enough to produce a crop such as millet, groundnut or cowpea. During this limited time, *Meloidogyne* populations will increase, but in the subsequent eight months of drought without vegetation, the populations will decrease to a very low level. In general, cowpea is grown in rotation with sorghum, millet and groundnut, all poor hosts or non-hosts of *Meloidogyne*; therefore, it is cultivated on the same land four months every three or four years and the root-knot populations will barely maintain themselves on an occasional weed during this period. Thus, the populations will be too low to be of any consequence when this crop is grown again. Under conditions of constant irrigation, however, populations of *Meloidogyne* will build up rapidly and usually less than two years are required before farmers encounter difficulties growing crops in fields

continuously cropped. This was clearly illustrated at the Centre National de Recherches Agronomiques at Bambey, Senegal, where cowpeas grown under field conditions did not show any sign of parasitism by *Meloidogyne*, whereas vegetable gardens situated near these fields that were irrigated throughout the year were heavily infested (NETSCHER, unpublished).

Under conditions favourable for the development of *Meloidogyne* (continuous cropping of susceptible plants under irrigation) a root-knot problem appears sooner or later and once a population of *Meloidogyne* is established, it is virtually impossible to eliminate. Nematicides may protect a crop for about three months permitting a satisfactory yield, but cannot prevent reestablishment of the population starting from the nematodes that survived the treatment. Even in moderately infested fields the use of resistant tomatoes does not eliminate a root-knot nematode population (NETSCHER and MAUBOUSSIN, 1973). Therefore, every means should be applied to avoid establishment of *Meloidogyne* in newly cultured land and to keep populations low in infested land. To achieve this goal, a truly integrated control system should be developed in which non-hosts, resistant varieties, cultural measures and nematicides should have each a place, keeping in mind the physiological variability, and thus the pathogenic capabilities of these parasites.

5. SUMMARY

The extreme morphological and physiological variability of certain root-knot nematodes (*Meloidogyne* spp.) and its implication on the development of control methods of these parasites, based upon crop rotations and the use of resistant varieties of otherwise susceptible crops are discussed.

In a review of the systematics of the genus *Meloidogyne* reestablished in 1949 by Chitwood to replace the polyphagous species *Heterodera marioni* Cornu, 1887, the variability of the so-called perineal pattern of the females, the most important character to distinguish species within the genus, is emphasized.

The results of the cytological investigations of Triantaphyllou are briefly discussed. It has been shown that most species studied are characterized by a parthenogenetic mode of reproduction. Two types of non-amphimictic reproduction have been observed: mitotic and meiotic parthenogenesis. *Meloidogyne incognita*, *M. javanica* and *M. arenaria* are characterized by mitotic parthenogenesis.

An analysis of the number of publications appearing between 1949 and 1976, dealing with identified species of *Meloidogyne* has revealed that 93% of the articles concern Chitwood's species of 1949, and that 76% refer to *M. incognita*, *M. javanica* and *M. arenaria*.

Faunistic studies in West Africa have shown that the most frequently encountered root-knot nematodes belong to the species *M. incognita*, *M. javanica* and *M. arenaria*. Identification of West African populations are complicated by the occurrence of mixtures of species, the great morphological variability and the existance of populations possessing rather large proportions of individuals exhibiting characters intermediate between species.

Certain morphometric characters of perineal patterns (width of vulva and distance between phasmids) were measured in a number of clones of *Meloidogyne* populations. These observations have demonstrated that although these characters are stable within clones, they cannot distinguish *M. javanica* from *M. incognita*.

The distance of the excretory pore of females from the anterior end, expressed in stylet lengths might possibly be an aid to distinguish *M. incognita* from *M. arenaria* and *M. javanica*.

The author is reluctant to describe populations characterized by unusual perineal patterns as new species and suggests inclusion of host-plant and cytological data in the description of new species.

Data presented indicate that length of juveniles could not be used to distinguish among *M. incognita*, *M. javanica* and *M. arenaria*.

Observations of juveniles from a population of *M. incognita acrita* have demonstrated that the inflation of the rectum is not an absolute criterium to distinguish *M. incognita* and *M. acrita*. A critical evaluation of the data of TERENTEVA (1967) concerning the height of the lip region of males of *M.*

incognita and *M. incognita acrita* makes the author reject the statement that this character might be useful to distinguish between these two taxa.

The author concludes that up till now, no infallible methods have been found to identify naturally occurring populations of *Meloidogyne* in West Africa.

After the splitting of the polyphagous *Heterodera marioni* into a number of species of *Meloidogyne* it became possible to assemble host lists for each species. In principal this information should provide a basis for crop rotation recommendations, intended to reduce root-knot nematodes infestation, provided that the *Meloidogyne* populations could be identified to the species level. Comparison of the host ranges of *M. incognita*, *M. javanica* and *M. arenaria* emphasized that these species have a great number of host plants in common, many of which are important crops. Only a few species could be used as differential plants to distinguish among these three species.

Published data and original results presented show that when many populations of the same species of *Meloidogyne* are studied, resistance/susceptibility of a given plant species cannot be predicted with confidence. Certain populations are able to parasitize a given plant and others are not.

The same phenomenon is observed with resistant varieties of otherwise susceptible crops. When several populations of the same species of *Meloidogyne* are tested against such a variety, those populations capable of parasitizing the resistant varieties are often called 'B races'. Studies concerning development of 'B races' on resistant tomatoes in Senegal have shown that some populations of *Meloidogyne* are able to parasitize resistant varieties strongly and immediately, others are able to form 'B races' after a selection has taken place; the majority however, is not capable of parasitizing resistant tomatoes.

Indications have been obtained that a gene for gene relation exists between nematode populations and resistant tomato varieties and a code indicating nematode and resistant variety genotype is proposed.

It is assumed that most plant species are attacked by *Meloidogyne* populations; the reaction of the plant attacked towards the specific *Meloidogyne* population concerned, determines if a successful parasitic relation develops.

It is proposed to consider as a group the tropical polyphagous species of *Meloidogyne*, which are characterized by a certain degree of polyploidy and a mitotic parthenogenetic mode of reproduction. This group should comprise *M. incognita*, *M. javanica*, *M. arenaria*, populations intermediate between these species, possibly undescribed species and certain described species for which information on mode of reproduction and physiological characters (host-range) is lacking.

On the basis of an actual field trial in Senegal, it is shown that recommendations for crop sequences or rotations should be based on testing different crops and varieties against naturally occurring populations of root-knot nematodes. Incorrect choice of crops may be made if rotations are based upon *Meloidogyne* identifications only. It is recommended that different populations in an area be tested on cash, food, and cover crops and resistant varieties.

Selection of resistant varieties should be based on reactions to as many

different populations of *Meloidogyne* as possible. Using such a technique 'B races' may be detected, and in addition much material not possessing root-knot resistance in the area where the varieties are developed, may be found to have resistance against other populations elsewhere.

In order to obtain the maximum value of non-hosts and resistant varieties, it is recommended to use these plants as a preventive measure rather than as a cure. Therefore, their use should be recommended in slightly infested or *Meloidogyne*-free soils.

The author has observed in fact, that non-hosts planted in heavily infested soil may be badly damaged because of a necrotic reaction to invading juveniles. In extreme cases this may give rise to dead patches in the field and a resultant increase of weed growth, accompanied by an increase of *Meloidogyne* on susceptible weeds. Thus, usefulness of the non-host can be nullified. Moreover the risk of selecting 'B races' is much higher in heavily infested land than in non- or slightly infested fields.

Thus it is recommended to use non-hosts and resistant varieties as preventive treatments within an integrated control including the various chemical and physical treatments available.

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7. SAMENVATTING

De buitengewoon grote morphologische en physiologische variabiliteit van sommige wortelknobbelaaltjes (*Meloidogyne* spp.) en de gevolgen hiervan op het ontwikkelen van bestrijdingsmethoden gebaseerd op vruchtwisseling en het gebruik van resistente rassen van overigens vatbare gewassen, worden besproken.

In een overzicht van de systematiek van het ter vervanging van de polyphage soort *Heterodera marioni* CORNU, 1887 door Chitwood in 1949 her-ingevoerde geslacht *Meloidogyne*, wordt vooral de nadruk gelegd op de van meet af aan geconstateerde variabiliteit van het zogenaamde perineaal patroon van de wijfjes, het belangrijkste kenmerk om de soorten binnen dit geslacht te onderscheiden.

De resultaten van het onderzoek van Triantaphyllou worden in het kort besproken. Aangetoond is dat de meeste tot nu toe onderzochte soorten gekenmerkt worden door een parthenogetische voortplanting. Twee typen van niet-geslachtelijke voortplanting zijn waargenomen: meiotische en mitotische parthenogenese. *Meloidogyne incognita*, *M. javanica* en *M. arenaria* worden gekenmerkt door een mitotische parthenogenese.

Een analyse van publicaties verschenen tussen 1949 en 1976, welke gedetermineerde *Meloidogyne* soorten als onderwerp hebben, heeft aangetoond dat in 93% van de artikelen Chitwood's in 1949 beschreven soorten worden behandeld, 75% van de artikelen hebben betrekking op *M. incognita*, *M. javanica* en *M. arenaria*.

Faunistische studies in West Afrika hebben aangetoond dat de meest voorkomende wortelknobbelaaltjes behoren tot de soorten *M. incognita*, *M. javanica* en *M. arenaria*.

Determinatie van de West-Afrikaanse populaties wordt bemoeilijkt door het veelvuldig samen voorkomen van meer dan een soort, de grote morphologische variabiliteit en het bestaan van populaties binnen welke individuen voorkomen, gekenmerkt door intermediaire kenmerken tussen twee soorten.

Enkele morphometrische kenmerken van perineaal patronen (breedte van de vulva en afstand tussen phasmiden) werden bepaald bij een aantal clonen van *Meloidogyne* populaties. Deze waarnemingen hebben aangetoond dat binnen clonen deze kenmerken weinig variëren, maar dat zij niet geschikt zijn om *M. javanica* en *M. incognita* te onderscheiden.

De afstand van de excretie-porie tot de voorkant van het lichaam van de wijfjes uitgedrukt in stekellengten, kan misschien een hulpmiddel zijn om *M. incognita* van *M. javanica* en *M. arenaria* te onderscheiden.

De schrijver zou willen vermijden dat populaties welke gekenmerkt zijn door ongebruikelijke perineale patronen als nieuwe soorten worden beschreven: gegevens omtrent cytologie en waardplanten behoren opgenomen te worden in beschrijvingen van nieuwe *Meloidogyne* soorten.

Gegevens over de lengte van larven van een aantal West-Afrikaanse populaties van *Meloidogyne* zijn verzameld: aangetoond is dat dit kenmerk niet gebruikt kan worden om *M. incognita*, *M. javanica* en *M. arenaria* te onderscheiden.

Waarnemingen bij larven van *M. incognita acrita* tonen aan dat het al of niet gezwollen zijn van het rectum van larven geen absoluut geldig kenmerk is om *M. incognita* en *M. incognita acrita* te onderscheiden. Een kritische beschouwing van de gegevens van TERENTEVA (1967) omtrent de hoogte van de lipstreek van mannetjes van deze twee soorten doet de schrijver concluderen dat ook dit kenmerk ongeschikt is om deze twee soorten te onderscheiden.

Na het opsplitsen van de zeer polyphage soort *Heterodera marioni* in een aantal *Meloidogyne* soorten was het mogelijk waardplantenlijsten voor iedere *Meloidogyne* soort samen te stellen. Deze kennis zou benut kunnen worden om vruchtwisselingen te ontwikkelen, bestemd om *Meloidogyne* populaties in besmette gronden te verminderen, op voorwaarde dat de in de grond aanwezige populaties met zekerheid gedetermineerd kunnen worden. Bij het samenstellen van waardplanten lijsten voor de verschillende *Meloidogyne* soorten bleek al spoedig dat *M. incognita*, *M. javanica* en *M. arenaria* een groot aantal waardplanten, waaronder veel landbouwgewassen, gemeen hadden. Gepubliceerde en eigen gegevens tonen aan dat als veel populaties van dezelfde *Meloidogyne* soort worden bestudeerd, de resistentie en/of vatbaarheid van een gegeven plantensoort niet met zekerheid kan worden voorspeld. Sommige populaties zullen in staat zijn een bepaalde plant te parasiteren en andere niet.

Hetzelfde verschijnsel is waargenomen bij resistente variëteiten van overigens vatbare gewassen. Als verscheidene populaties van dezelfde *Meloidogyne* soort worden getoetst op een voor deze soort resistente variëteit, worden die populaties welke in staat zijn deze plant te parasiteren wel 'B rassen' genoemd. Onderzoek omtrent de ontwikkeling van 'B rassen' op resistente tomaten in Senegal heeft aangetoond dat sommige *Meloidogyne* populaties in staat zijn resistente variëteiten onmiddellijk sterk aan te tasten; andere populaties kunnen 'B rassen' vormen nadat een selectie heeft plaats gevonden; de meerderheid is echter niet in staat resistente tomaten te parasiteren.

Aangenomen wordt dat de meeste plantensoorten worden aangevallen door *Meloidogyne* soorten en dat het van de reactie van de aangetaste planten afhangt of de nematoden zich zullen ontwikkelen.

Voorgesteld wordt om de belangrijkste tropische, polyphage *Meloidogyne* soorten te groeperen. Deze groep wordt gekenmerkt door polyploide soorten met een mitotisch parthenogenetische voortplanting en omvat *M. incognita*, *M. javanica* en *M. arenaria*, intermediaire populaties, misschien nog onbeschreven soorten en mogelijk bestaande soorten waarvan gegevens omtrent voortplanting en physiologische kenmerken (waardplantenlijsten) ontbreken.

Op grond van een veldproef in Senegal wordt aangeraden aanbevelingen voor vruchtwisselingen te baseren op toetsingen van wortelknobbelaaltjes op verschillende gewassen en variëteiten. Een foutieve keuze van gewassen kan gemaakt worden als vruchtwisselingen worden voorgesteld naar aanleiding van

determinatie van *Meloidogyne* populaties. Aanbevolen wordt verschillende populaties binnen een gebied te toetsen op handelsgewassen, voedselgewassen, bodembedekkers en resistente variëteiten.

Veredeling op resistentie moet gebaseerd worden op het toetsen van zo veel verschillende *Meloidogyne* populaties als technisch mogelijk is. Op deze wijze kunnen 'B rassen' ontdekt worden; bovendien kunnen planten die geen resistentie vertonen in het gebied waar de selectie plaats vindt elders waardevol zijn.

Ten einde zo veel mogelijk te profiteren van niet door *Meloidogyne* geparasiteerde plantensoorten en resistente variëteiten moeten deze in preventieve en niet als curatieve methoden worden benut. Aangeraden wordt deze planten te gebruiken in weinig of niet besmette gronden. De schrijver heeft waargenomen dat niet door *Meloidogyne* geparasiteerde plantensoorten, verbouwd in zwaar besmette grond ernstig beschadigd kunnen worden door necrose als reactie op in de plant binnengedrongen larven. In extreme gevallen kunnen planten plekksgewijze afsterven hetgeen een sterke ontwikkeling van onkruiden, waaronder vele waardplanten van *Meloidogyne*, met zich mee brengt. In zulke gevallen zal het overigens gunstige effect van het gekozen resistente gewas te niet worden gedaan. Ook verhoogt de teelt van resistente variëteiten in zwaar besmette gronden het gevaar van selectie van 'B rassen'.

Een geïntegreerde bestrijding waarin chemische en physische bestrijdingsmethoden hun plaats hebben naast vruchtwisselingen met onvatbare gewassen en resistente variëteiten wordt gezien als de beste oplossing van het *Meloidogyne* probleem.

8. REFERENCES

ADDOH, P. G. (1970). The root-knot nematode problem in Ghana: host and non-host plants of *Meloidogyne* species. Ghana Jnl. agric. Sci. **3**: 3–12.

ALLEN, M. W. (1952). Observations on the genus *Meloidogyne* Goeldi, 1887. Proc. helminth. Soc. Wash. **19**: 44–51.

BAKER, A. D. (1953). Rapid method for mounting nematodes in glycerine. Can. Ent. **85**: 77–78.

BAKER, A. D. (1962). Check lists of the nematode superfamilies Dorylaimoidea, Rhabditoidea, Tylenchoidea, and Aphelenchoidea. E. J. Brill, Leiden, Netherlands: 261 pp.

BARKER, K. R., SHOEMAKER, P. B. & NELSON, L. A. (1976). Relationships of initial population densities of *Meloidogyne incognita* and *M. hapla* to yield of tomato. J. Nematol. **8**: 232–239.

BARRONS, K. C. (1939). Studies of the nature of root knot resistance. J. agric. Res. **58**: 263–271.

BIRD, A. F. (1974). Plant response to root-knot nematode. A. Rev. Phytopathol. **12**: 69–85.

BRUN, J. L. (1966). L'adaptation aux températures élevées chez un nématode: *Caenorhabditis elegans* Maupas, 1900. Rôle des facteurs autres que la température dans le processus d'adaptation. Ann. Biol. anim. Biochem. Biophys. **6**: 439–466.

CAVENESS, F. E. (1962). Nematology in Western Nigeria. End of tour progress report of the nematology project. Ministry of Agriculture and Natural Resources, Western Region, Nigeria; United States of America. Agency for International Development Project 61-13-050: 60 p.

CHITWOOD, B. G. (1949). 'Root knot nematodes' – Part I. A revision of the genus *Meloidogyne* Goeldi, 1887. Proc. helminth. Soc. Wash. **16**: 90–104.

CHITWOOD, B. G. & OTEIFA, B. A. (1952). Nematodes parasitic on plants. A. Rev. Microbiol. **6**: 151–184.

CHITWOOD, B. G., SPECHT, A. W. & HAVIS, L. (1952). 'Root-knot nematodes. III. Effects of *Meloidogyne incognita* and *M. javanica* on some peach rootstocks.' Pl. & Soil **4**: 77–95.

CHRISTIE, J. R. (1949). Host parasite relationship of the root-knot nematode, *Heterodera marioni*, III. The nature of resistance in plants to root-knot. Proc. helminth. Soc. Wash. **16**: 104–108.

CHRISTIE, J. R. & ALBIN, F. E. (1944). Host-parasite relationships of the root-knot nematode, *Heterodera marioni*. I. The question of races. Proc. helminth. Soc. Wash. **11**: 31–37.

COLBRAN, R. C. (1958). Studies of plant and soil nematodes. 2. Queensland host records of root-knot nematodes (*Meloidogyne* species). Qd J. agric. Sci. **15**: 101–136.

DALMASSO, A. & BERGÉ, J. B. (1975). Variabilité génétique chez les *Meloidogyne* et plus particulièrement chez *M. hapla*. Cah. ORSTOM, Sér. Biol. **X**: 233–238.

DE GUIRAN, G. (1960). Étude comparative de la pénétration des larves de *Meloidogyne javanica* (Treub, 1885) Chitwood, 1949 et de *Meloidogyne incognita acrita* Chitwood, 1949, dans les racines des plantes hôtes et non hôtes. Résultats préliminaires. Meded. LandbHoogesch. OpzoekStns Gent. **25**: 1047–1056.

DROPKIN, V. H. (1953). Studies on the variability of anal plate patterns in pure lines of *Meloidogyne* spp., the root-knot nematode. Proc. helmith. Soc. Wash. **20**: 32–39.

DROPKIN, V. H. (1969). The necrotic reaction of tomatoes and other hosts resistant to *Meloidogyne*: Reversal by temperature. Phytopathology **59**: 1632–1637.

ESSER, R. P., PERRY, V. G. & TAYLOR, A. L. (1976). A diagnostic compendium of the genus *Meloidogyne* (Nematoda: Heteroderidae). Proc. helminth. Soc. Wash. **43**: 138–150.

FENWICK, D. W. & FRANKLIN, M. T. (1942). Identification of *Heterodera* species by larval length. Technique for estimating the constants determining the length variations within a given species. J. Helminth. **20**: 67–114.

FRANKLIN, M. T. (1971). Taxonomy of Heteroderidae. *In:* Plant parasitic nematodes. Edited

by B. M. Zuckermann, W. F. Mai and R. A. Rohde. Academic Press, New York and London, Vol. I: 139–162.
GIAMALVA, M. J., MARTIN, W. J. & HERNANDEZ, T. P. (1963). Sweet potato varietal reaction to species and races of root-knot nematodes. Phytopathology **53**: 1187–1189.
GIEBEL, J. (1974). Biochemical mechanisms of plant resistance to nematodes: A review. J. Nematol. **6**: 175–184.
GOLDEN, A. M. (1974). *Meloidogyne incognita*: a single, homogeneous species or a complex of two or more taxa? J. Nematol **6**: 141.
GOLDEN, A. M. (1976). A review of the basic aspects of the genus *Meloidogyne*. *In*: Proceedings of the Research Planning Conference on root-knot nematodes, *Meloidogyne* spp. January 12–16, N. C. State University, Raleigh.
GOODEY, J. B., FRANKLIN, M. T. & HOOPER, D. J. (1965). T. Goodey's The nematode parasites of plants catalogued under their hosts. Commonwealth Agricultural Bureaux, Farnham Royal, Bucks, England: 214 p.
HACKNEY, R. W. (1974). The use of chromosome number to identify *Meloidogyne* spp. on grapes. J. of Nematol. **6**: 141–142.
HINSHELWOOD, C. N. (1947). The chemical kinetics of the bacterial cell. Clarendon Press, Oxford: 284 p.
IBRAHIM, I. K. A. & EL-SAEDY, M. A. (1976). Development and parthenogenesis of *Meloidogyne javanica* in peanut roots. Nematol. medit. **4**: 231–234.
JATALA, P. & RUSSELL, C. C. (1972). Nature of sweet potato resistance to *Meloidogyne incognita* and the effects of temperature on parasitism. J. Nematol. **4**: 1–7.
KEHR, A. E. (1966). Current status and opportunities for the control of nematodes by plant-breeding. *In*: Pest control by chemical, biological, genetic and physical means. A symposium. Agricultural Research Service. United States Department of Agriculture.
KIRBY, M. F., DICKSON, D. W. and SMART, G. C. (1975). Physiological variation within species of *Meloidogyne* occurring in Florida. Plant. Dis. Reptr **59**: 353–356.
MARTIN, G. C. (1956). The common root-knot nematode (*Meloidogyne javanica*) and crop rotation. 5. Cotton (*Gossypium hirsutum* var. 9L34). Rhod. Fmr **27**: 33.
MARTIN, W. J. & BIRCHFIELD, W. (1973). Further observations of variability in *Meloidogyne incognita* on sweet potatoes. Pl. Dis. Reptr **57**: 199.
MERNY, G. & CADET, P. Penetration of juveniles and development of adults of *Heterodera oryzae* on several hosts and non hosts. (in press).
MILNE, D. J. (1972). Nematodes of tobacco. *In*: Economic Nematology. Edited by J. M. Webster. Academic Press, London and New York: 159–186.
MINTON, N. A. (1963). Effects of two populations of *Meloidogyne arenaria* on peanut roots. Phytopathology **63**: 79–81.
MINTON, N. A., MCGILL, J. F. & GOLDEN, A. M. (1969). *Meloidogyne javanica* attacks peanuts in Georgia. Pl. Dis. Reptr **53**: 668.
MINZ, G. (1958). *Meloidogyne javanica* in strawberry roots. Pl. Prot. Bull. F.A.O. **6**: 92.
NETSCHER, C. (1970). a. Les nématodes parasites des cultures maraîchères au Sénégal. Cah. ORSTOM, Sér. Biol. **11**: 209–229.
NETSCHER, C. (1970). b. A rapid technique for mass-killing of nematodes with hot fixative. Nematologica **16**: 603.
NETSCHER, C. (1973). Étude sur la variabilité de la longueur des larves chez *Meloidogyne incognita* Chitwood, 1949, et *Meloidogyne javanica* Chitwood, 1949. Cah. ORSTOM, Sér. Biol. **21**: 91–95.
NETSCHER, C. (1974). L'arachide et le contrôle biologique des nématodes *Meloidogyne* spp. dans les cultures maraîchères du Sénégal. C. r. hebd. Séanc. Acad. Agric. Fr. **60**: 1332–1339.
NETSCHER, C. (1975). Studies on the resistance of groundnut to *Meloidogyne* sp. in Senegal. Cah. ORSTOM, Sér. Biol. **X**: 227–232.
NETSCHER, C. (1977). Observations and preliminary studies on the occurrence of resistance breaking biotypes of *Meloidogyne spp.* on tomato. Cah. ORSTOM, Sér. Biol. **XI**, 1976: 173–178.

NETSCHER, C. & LUC, M. (1974). Nématodes associés aux cultures maraîchères en Mauritanie. Agron. trop., Nogent **29**: 697–701.
NETSCHER, C. & MAUBOUSSIN, J. C. (1973). Résultats d'un essai concernant l'efficacité comparée d'une variété de tomate résistante et de certains nématicides contre *Meloidogyne javanica*. Cah. ORSTOM, Sér. Biol. **21**: 97–102.
NETSCHER, C. & PERNES, J. (1971). Étude concernant l'influence de la constitution génétique sur la longueur des larves d'*Heterodera oryzae*. Nematologica **17**: 336–346.
NETSCHER, C. & SEINHORST, J. W. (1969). Propionic acid better than acetic acid for killing nematodes. Nematologica **15**: 286.
OOSTENBRINK, M. (1957). Das vorkommen von Artgemischen bei pflanzenparasitären Nematoden. Nematologica **2**, Suppl.: 342–346.
OTEIFA, B. A., ELGINDI, D. M. & MOUSSA, F. F. (1970). Root-knot problem in recently reclaimed sandy areas of U.A.R. I. Nematode infestation and host range. Meded. Fak. LandbWeten. Gent **35**, 1167–76.
PERRY, V. G. & ZEIKUS, J. A. (1972). Host variations among populations of the *Meloidogyne incognita* group. J. Nematol. **4**: 231–232.
PHIPPS, P. M., STIPES, R. J. & MILLER, L. I. (1972). A race of *Meloidogyne incognita* from *Albizzia julibrissin* parasitizes *Nicotiana tabacum* 'N.C. 95'. J. Nematol. **4**: 232.
RASKI, D. J., ALLEN, M. W. & BURTON, V. E. (1953). Nematode on cotton. Root-knot nematode control by soil fumigation profitable in Kern county. Calif. Agric. **7**: 8–13.
REYNOLDS, H. W., CARTER, W. C. & O'BANNON, J. H. (1970). Symptomless resistance of alfalfa to *Meloidogyne incognita acrita*. J. Nematol. **2**: 131–138.
RIGGS, R. D. & WINSTEAD, N. N. (1959). Studies on resistance in tomato to root-knot nematodes and on the occurrence of pathogenic biotypes. Phytopathology **49**: 716–724.
SANDERS, H. & MULET, J. (1976). 'E' type perineal patterns in *Meloidogyne* species. Nematologica **22**: 373–376.
SASSER, J. N. (1954). Identifications and host-parasite relationships of certain root-knot nematodes (*Meloidogyne* spp.). Bull. Md agric. Exp. Stn A-77: 31 p.
SASSER, J. N. (1966). Behavior of *Meloidogyne* spp. from various geographical locations on ten host differentials. Nematologica **12**: 97–98.
SASSER, J. N. (1977). The distribution of the root-knot nematodes in the potato-production areas of the world. Organization of Tropical American Nematologists (OTAN). Annual meeting Lima, Peru, March 1977: 16 p.
SAUER, M. R. & GILES, J. E. (1959). A field trial with a root-knot resistant tomato variety. C.S.I.R.O. Austr. Irrig. Res. Sta., Tech. Pap. **3**: 1–10.
SEINHORST, J. W. (1959). A rapid method for the transfer of nematodes from fixative to anhydrous glycerin. Nematologica **4**: 67–69.
SIDHU, G., WEBSTER, J. M. (1974). Genetic control of resistance in tomato. I. Identification of genes for host resistance to *Meloidogyne incognita*. Nematologica **19**, 1973: 546–550.
SIHIDU, G. & WEBSTER, J. M. (1974). Genetic control of resistance in tomato. I. Identification of resistance in tomato (*Lycopersicon esculentum*) against *Meloidogyne incognita*. Can. J. Genet. Cytol. **17**: 323–328.
SIMPSON, G. G. (1961). Principals of animal taxonomy. Columbia University Press, New York: 247 p.
SITTERLY, W. R. & FASSULIOTIS, G. (1965). Potato losses in South Carolina due to the cotton root-knot nematode, *Meloidogyne incognita acrita*. Pl. Dis. Reptr **49**: 723.
SLEDGE, E. B. & GOLDEN, A. M. (1964). *Hypsoperine graminis* (Nematoda: Heteroderidae), a new genus and species of plant-parasitic nematode. Proc. helminth. Soc. Wash. **31**: 83–88.
SOUTHARDS, C. J. & PRIEST, M. F. (1973). Variation in pathogenicity of seventeen isolates of *Meloidogyne incognita*. J. Nematol. **5**: 63–67.
TAKA, A. H. Y. & YOUSIF, G. M. (1976). Histopathology of peanut underground parts infected with *Meloidogyne incognita*. Nematol. medit. **4**: 175–181.
TAYLOR, A. L., DROPKIN, V. H. & MARTIN, G. C. (1955). Perineal patterns of root-knot nematodes. Phytopathology **45**: 26–34.

TAYLOR, D. P. (1976). Plant nematology problems in tropical Africa. Helminthol. Abstr., B. **45**: 269–284.
TAYLOR, D. P. & NETSCHER, C. (1974). An improved technique for preparing perineal patterns of *Meloidogyne* spp. Nematologica **20**: 268–269.
TAYLOR, D. P. & NETSCHER, C. (1975). Occurrence in Senegal of a biotype of *Meloidogyne javanica* parasitic on strawberry. Cah. ORSTOM, Sér. Biol. **X**: 247–249.
TERENTEVA, T. G. (1967). Use of statistics for testing the variability of taxonomic characters of species of *Meloidogyne*. *In*: Nematode diseases of crops. Edited by N. M. Svesnikova: Izdatelstvo 'Kolos', Moscow: 223–227. (In Russian).
TOXOPEUS, H. J. (1956). Some remarks on the development of new biotypes in *Heterodera rostochiensis* that might attack resistant potato-clones. Nematologica **1**: 100–101.
TRIANTAPHYLLOU, A. C. (1962). Oogenesis in the root-knot nematode *Meloidogyne javanica*. Nematologica **7**: 105–113.
TRIANTAPHYLLOU, A. C. (1963). Polyploidy and parthenogenesis in the root-knot nematode *Meloidogyne arenaria*. J. Morph. **113**: 489–499.
TRIANTAPHYLLOU, A. C. (1966). Polyploidy and reproductive patterns in the root-knot nematode *Meloidogyne hapla*. J. Morph. **118**: 403–414.
TRIANTAPHYLLOU, A. C. (1969). Gametogenesis and the chromosomes of two root-knot nematodes, *Meloidogyne graminicola* and *M. naasi*. J. Nematol. **1**: 62–71.
TRIANTAPHYLLOU, A. C. (1971a). Gametogenesis and reproduction of *Meloidogyne graminis* and *M. ottersoni* (Nematoda: Heteroderidae). J. Nematol. **5**: 84–87.
TRIANTAPHYLLOU, A. C. (1971b). Genetics and cytology. *In*: Plant parasitic nematodes. Edited by B. M. Zuckermann, W. F. Mai and R. A. Rohde. Acad. Press Inc., New York and London, Vol. **2**: 347 p.
TRIANTAPHYLLOU, A. C. & SASSER, J. N. (1960). Variation in perineal patterns and host specifity of *Meloidogyne incognita*. Phytopathology **50**: 724–735.
USTINOV, A. A. (1959). The gall nematode. Kharkov: Izdatelstvo Kharkovskogo Gosudarstvennogo Universiteta: 293 p. (In Russian).
WHITEHEAD, A. G. (1968). Taxonomy of *Meloidogyne* (Nematoda: Heteroderidae) with description of four new species. Trans. zool. Soc. London **31**: 267–401.
WHITEHEAD, A. G. (1969). The distribution of root-knot nematodes (*Meloidogyne* spp.) in tropical Africa. Nematologica **15**: 315–333.
WINOTO SUATMADJI, R. (1969). Studies of the effect of *Tagetes* species on plant parasitic nematodes. Veenman & Zonen, N.V., Wageningen, The Netherlands: 132 p.
WOUTS, W. M. and SHER, S. A. (1971). The genera of the subfamily Heteroderinae (Nematoda: Tylenchoidea) with a description of two new genera. J. Nematol. **3**: 129–144.

CURRICULUM VITAE

Caspar Netscher werd op 21 juni 1931 geboren te 's-Gravenhage. Na het behalen van het einddiploma van de Rijks H.B.S. te 's-Hertogenbosch studeerde hij aan de Landbouwhogeschool te Wageningen, waar op 24 september 1960 het diploma Landbouwkundig Ingenieur werd behaald.

Van october 1960 tot juni 1962 was hij verbonden aan de University of Science and Technology te Kumassi (Ghana) als phytopatholoog.

In augustus 1962 werd hij als nematoloog aangesteld bij het ORSTOM (Office de la Recherche Scientifique et Technique Outre-Mer), waar hij nog steeds werkzaam is. Tot juni 1970 werkte hij in het Laboratorium voor Nematologie van deze organisatie te Adiopodoumé (Côte d'Ivoire) waarna dezelfde functie werd vervuld in het Laboratorium voor Nematologie te Dakar (Senegal).